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# ARCHIVES OF PATHOLOGY

VOLUME 18

JULY 1934

NUMBER 1

## EXTRAMEDULLARY ERYTHROCYTOPOIESIS IN MAN

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Extramedullary hemopoiesis is a common phenomenon associated with certain anemias and leukemias. The condition is generally interpreted as compensatory for a hypofunctional bone marrow. Uncertainty concerns especially the source of the proerythrocyte in the metaplastic tissues. It is the purpose of this study to describe in detail two cases of extensive erythroid metaplasia, and to attempt an interpretation of the special conditions in the light of the data revealed by the evolutionary history of blood-forming tissues. Such interpretations must recognize the fundamental importance of the spleen in the production of the red cells and the potential erythrocytogenic capacity of the lymphocytes.

The two cases of erythroid metaplasia under consideration are complementary in that one shows extensive involvement of lymph nodes following damage to the bone marrow and spleen while the other shows very active splenic hemocytopenia associated with a considerable replacement of lymph nodes and bone marrow by metastatic adenocarcinoma from the prostate.

### CASE 1<sup>1</sup>

#### CLINICAL HISTORY

A white woman of about 40 years was admitted to the University Hospital on June 23, 1924. She complained of weakness and of a mass in the left side. She first noticed the mass about one year earlier. It had progressively grown larger, particularly since January, 1924. She presented a perplexing condition indicative of leukemia, but whether this was myelocytic, lymphatic or mixed was uncertain. She grew rapidly worse. The very large spleen receded after treatment with radium on June 28, but the blood picture did not improve. The exposure to radium of 3,200 mg.-hours led to a rather violent reaction. She died unexpectedly on July 8, 1924.

On admission the erythrocytes numbered 1,050,000 per cubic millimeter; on July 4, six days after treatment with radium, 886,000. On admission, the hemoglobin (Dare) was 15 per cent. The bleeding time was twenty-six minutes; the clotting time, six minutes. The platelets numbered 39,000. The leukocyte count was 70,000; two days after treatment with radium, 9,200; four days later, 3,200. The differential count on admission was: neutrophils, 10.6 per cent; small lymphocytes,

From the Laboratory of Histology and Embryology, University of Virginia.

1. The materials studied in this case are on file with the Lymphatic Tumor Registry as Accession No. 32825. A brief report was published in collaboration with Dr. H. T. Marshall under the title, "Metaplastic Development of Erythrocytes in Lymph Nodes," Anat. Rec. 29:363, 1925.

83.3 per cent; large lymphocytes, 3.7 per cent; eosinophils, 0.3 per cent; mast cells, 0.1 per cent; transitionals, none; neutrophilic myelocytes, 1.4 per cent; eosinophilic myelocytes, 0.5 per cent; basophilic myelocytes, 0.1 per cent. Two normoblasts and 1 microblast were seen.

#### AUTOPSY REPORT

The condition of the lungs suggested early pneumonia. The peribronchial lymph nodes were large, moist, congested and ecchymotic, and soft. The immediate cause of death was probably bronchopneumonia of hemorrhagic type. Accompanying the pneumonia was a lymphadenitis of hemorrhagic type along the bronchial nodes. The spleen weighed 690 Gm.; its length was 17.5 cm.; its breadth about one half the length. The bone marrow of the femur was hemorrhagic and rather watery in texture, with small, spongy bits of tissue in the watery red background. There was no white marrow or fat. The bone marrow of the ribs and vertebrae was of similar character. The lymph nodes of the neck, axillae, mesentery and inguinal region were soft, somewhat enlarged and red or pink.

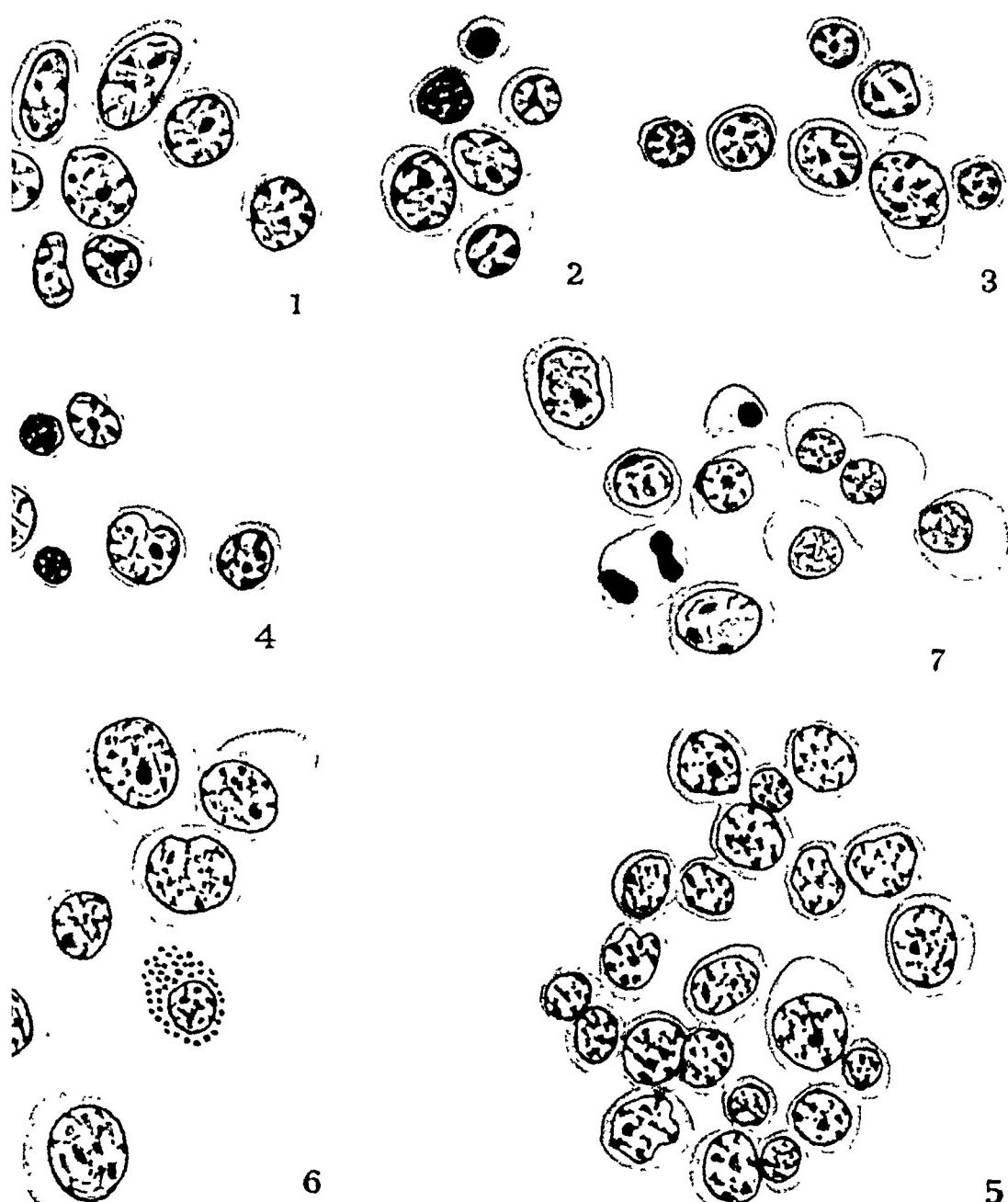
#### HEMOPOIESIS

*Marrow.*—Smears were made from a rib, a femur and a vertebra, and stained with Wright's stain. The small lymphocytes predominated; a considerable number of lymphocytes of intermediate size also occurred. Typical hemocytoblasts, erythroblasts and normoblasts were abundant. Only one megakaryocyte was seen. Granulocytes, both neutrophilic and eosinophilic, were rare. The absence of mitoses seems to indicate abeyance of local histogenetic activity.

Sections of femoral marrow, fixed in Helly's fluid<sup>1a</sup> and stained with either hematoxylin and eosin or the eosin-azure combination of Giemsa, gave the appearance of lymphoid metaplasia. Approximately half of a transverse section consisted of moderately dense lymphoid tissue, with a suggestion of several nodules. Small lymphocytes greatly predominated. A few large lymphocytes (hemocytoblasts) and numerous transitionals in the form of medium-sized lymphoid hemoblasts occurred (fig. 5). Many of the small lymphocytes were in stages of degeneration, as indicated by an extensive karyorrhexis. In the other half of the section definitive red blood corpuscles predominated; this region also contained widely scattered small and intermediate-sized lymphocytes and hemocytoblasts (fig. 6), erythrocytes at various stages of maturation (fig. 7), megakaryocytes and a few macrophages with erythrocyte débris. The area of junction between the compact lymphoid tissue and the loose erythroid tissue had an intermediate histologic structure. There was no indication of erythrocytogenic activity on the part of the endothelium.

- 
- 1a. The formula for Helly's fluid is as follows:

Potassium bichromate.....	2.5 Gm.
Sodium sulphate.....	1.0 Gm.
Mercuric chloride .....	5.0 Gm.
Distilled water.....	100.0 Cc.
Dilute formaldehyde solution.....	5.0 Cc.



#### EXPLANATION OF PLATE 1 (CASE 1, LEUKEMIA)

All of the drawings were made at a magnification of 1,350 diameters, from sections of tissues fixed in Helly's fluid, the sections having been stained with hematoxylin and eosin.

Fig. 1.—Group of cells from a splenic nodule. Included among them are small and large lymphoid hemoblasts and one erythroblast (below, at left). The large lymphoid hemoblast is the equivalent of the hemocytoblast; the small, of the lymphocyte.

Fig. 2.—Group of cells from splenic pulp. Included among them are two lymphoid hemoblasts, three erythroblasts and one normoblast.

Fig. 3.—Group of cells from the periphery of a nodule of a lymph node, including one large and six smaller lymphoid hemoblasts.

Fig. 4.—Group of cells from a sinus of a lymph node, including a large and two smaller lymphoid hemoblasts, an erythroblast and two normoblasts.

Fig. 5.—Group of lymphoid hemoblasts of various sizes from bone marrow. This region resembles compact lymphoid tissue.

Fig. 6.—Group of large lymphoid hemoblasts (hemocytoblasts) from a less dense area of bone marrow. The group includes one eosinophil. This area includes many erythroplastids and a few megakaryocytes.

Fig. 7.—Group of cells from a region intermediate between those of figures 5 and 6, including two hemocytoblasts, a number of erythroblasts (one in mitosis) and one normoblast.

*Spleen.*—Different regions of the spleen differed histologically. The difference inhered in relative abundance of lymphocytes and stroma. Over large areas, in the aggregate apparently through the greater volume, lymphocytes were almost entirely lacking. Here the stroma was very prominent, and the lining of the partially collapsed sinuses was composed of closely spaced rounded cells giving a false appearance of exfoliation into the lumen. There were, however, no areas of hyalinization. In other areas lymphocytes were fairly abundant, including both large (hemocytoblasts) and small varieties, the latter greatly in excess. Between these areas in which conditions were extreme there were areas of intermediate lymphocyte content (figs. 1 and 2). Occasional macrophages, laden with red cells at various stages of differentiation, including a lymphocyte or two, were to be found. Megakaryocytes were absent. Monocytes occurred sparingly. Nucleated red cells and granulocytes were relatively rare. The most abundant granulocyte was a spheronuclear basophil.

*Lymph Nodes.*—The lymph nodes were uniformly enlarged. The microscopic study included several each from the neck, axilla and groin. This tissue was fixed in Helly's mixture of Zenker solution and diluted formaldehyde solution, and was stained either with the eosin-azure combination of Giemsa or with hematoxylin and eosin. The most characteristic feature of the lymph nodes was an almost complete obliteration of the demarcation between the cortex and the medulla. The parenchyma consisted generally of a uniformly compact mass of small and medium-sized lymphocytes (fig. 3), resembling somewhat splenic pulp. Occasional more compact masses of small lymphocytes suggested a previous nodule. Such nodules generally had light-staining centers of connective tissue with evidence of beginning necrosis and hyalinization. Large lymphocytes (hemocytoblasts) were also abundant; a few were in mitosis. Mingled with the lymphocytes were large numbers of red cells at all stages of differentiation (fig. 4). Cells transitional between hemocytoblasts and normoblasts were present in large numbers. There was no evidence of endothelial activity in the formation of hemocytoblasts. Megakaryocytes were lacking. Occasional macrophages with erythrocyte débris occurred. Granulocytes were rare, basophils predominating.

The arterioles were generally empty, their walls contracted and relatively thick. The venules were generally engorged with lymphocytes and differentiating erythrocytes. The lymph nodes resembled splenic pulp, with large, apparently fenestrated sinusoid spaces. The differentiating red cells occurred both in these "pulp sinuses" and extravascularly among the lymphocytes. The appearance suggested an extravascular differentiation of red cells from lymphocytes. Mitoses occurred only in the large lymphocytes.

## COMMENT

An attempt at interpretation of the histology of the marrow, spleen and lymph nodes must take into account the cellular content of the peripheral blood. The outstanding differential feature was the drop in the number of leukocytes from 70,000 to 3,200 within six days after the treatment with radium. Neither the total red cell count of approximately 1,000,000, nor the relative proportions of the white cells were markedly affected by the radium treatment. The greatest difference relates to the small lymphocytes; these suffered a decrease in number, leaving a relative increase in the number of the neutrophilic granulocytes. Platelets were relatively rare, and eosinophilic and basophilic granulocytes were practically absent. Nucleated red cells were very scarce, one differential count revealing only 2 normoblasts. Only the granulocytes of these blood smears gave a positive oxydase reaction; the mononuclear leukocytes were uniformly negative.

I am not especially concerned here with an effort to classify this case of leukemia, whether lymphatic, myelogenous or "mixed"; it seems to conform most closely to the case described by Fineman<sup>2</sup> in which the condition was designated "microlymphoidocytic leukemia." Nor does it seem desirable here to review again the extensive literature dealing with the question of single, dual or multiple sources of "myelogenous" cells in leukemia. This literature has been very thoroughly reviewed by Logefeil.<sup>3</sup> I am concerned primarily with the fact that under certain pathologic conditions the lymph nodes may partially compensate functionally for a morbid bone marrow or spleen, and with the evidence that lymphocytes may differentiate into erythrocytes, and with the implications of these data with respect to the significance of the lymphocytes in normal hemocytogenesis.

Much of the recent work on leukemias leads to the conclusion that under the conditions prevailing in this type of disease the lymphocytes of the lymph nodes may differentiate into "myeloid cells." Thus Citron,<sup>4</sup> in his discussion of a case of "micromyeloblastic leukemia," stated his belief that "in some cases a direct autocellular change of lymphatic follicular lymphocytes into myeloid cells may take place" in both the spleen and lymph nodes. Fineman<sup>2</sup> in a case of microlymphoidocytic leukemia noted the abundance of forms transitional between lymphocytes and hemoblasts ("atypical cells") both in the peripheral blood and in the lymph nodes, but was unable to determine "whether the lymphocyte is the mother cell of our atypical cell or vice versa." Logefeil<sup>3</sup> found evidence in a case of "mixed leukemia" of a "direct transition from

2. Fineman, S.: Arch. Int. Med. **28**:168, 1922.

3. Logefeil, R. C.: Arch. Int. Med. **33**:659, 1924.

4. Citron, J.: Folia haemat. **20**:1, 1915.

lymphocytes to myelocytes, without going through the stage of the stem cell" (hemocytoblast). He noted also evidence of a local development of myelocytes from lymphocytes in the areas of leukemic infiltration in the pancreas, kidneys and lungs. Furthermore, in both the lymph nodes and the spleen he found "immature" lymphocytes and myelocytes diffusely arranged without evidence of segregation. He stated that "many of the eosinophils, both immature and adult, had nuclei identical with adjacent lymphocytes." He quoted Turk (1908) as having reported the presence of many nucleated red cells in the lymph nodes in a case of "mixed leukemia." But no definite statement appears as to whether these cells were believed to be the result of infiltration or of local differentiation. Finally, Downey<sup>5</sup> stated that "under pathological and experimental conditions which cause myeloid metaplasia the derivation of myeloid cells from lymphocytes without the intervention of the myeloblast may be an extensive process." He admitted further that the blood in lymphatic leukemia may show all transitional forms from the "myeloblast" (hemocytoblast) to the ordinary lymphocyte, without, however, committing himself as to which cell is the progenitor.

In the group of the Amphibia the evidence seems conclusive that the lymphocyte may function as a mother cell of both erythrocytes and granular leukocytes (Jordan<sup>6</sup>). This case of leukemia has especial interest because it furnishes apparently confirmatory evidence with respect to man. Here the data seem to exclude the possibility that the very numerous nucleated red cells of the lymph nodes were transported from the bone marrow. The peripheral blood contained very few nucleated red cells. The very numerous normoblasts could, therefore, not have entered the lymph nodes from the blood stream. Moreover, nucleated red cells, while numerous in the marrow, were relatively fewer there than in the lymph nodes. The erythroblastids apparently had origin largely in the lymph nodes but to some extent in the spleen and the marrow. Numerous stages in the enucleation of the normoblasts also appeared in the lymph nodes, spleen and marrow.

The possibility, of course, remains that the small lymphocyte-like cells of the lymph nodes, which differentiated here into erythrocytes, were specific proerythroblasts whose ancestors early in the morbid condition found lodgment in the lymph nodes, there to develop the erythrocytopoietic "metaplasia" of the leukemia. This objection cannot be met with any definite or final statement. It can only be said that on the basis of the morphologic and tinctorial criteria here possible of application no difference can be detected between alleged distinct lymphocyte ancestors and erythrocyte ancestors. The lymphoid cells of the lymph

5. Downey, H.: Arch. Int. Med. **33**:301, 1924.

6. Jordan, H. E.: Am. J. Anat. **51**:215, 1932.

nodes and the marrow which constituted the erythrocyte precursors in this case have an identical appearance in stained sections.

Briefly sketched the case may be interpreted as follows: The dysfunction of the marrow, the primary cause of which remains unknown, resulting chiefly in a condition of severe anemia, stimulated a compensatory reversion of the spleen to its embryonic hemocytopoietic condition. Relatively intense proliferative activity or inability of sufficiently rapid differentiation of splenic polyvalent lymphocytes into erythrocytes (due possibly, at least in part, to a lack of favorable conditions for the development of hemoglobin) caused an accumulation of the lymphocyte red-cell ancestors, with a consequent enlargement of the spleen. Radium irradiation then destroyed large numbers of the splenic lymphocytes with a consequent decrease in the size of the spleen and the production of the histologic condition of large areas of lymphocyte-free stroma. Following this intentional destruction of the lymphocytes of the spleen, compensation was attempted on the part of the only other available potentially myeloid tissue, namely, the lymph nodes.

## CASE 2

### MATERIALS AND METHODS

The materials of this study included the spleen, bone marrow, lymph nodes and liver in a case of adenocarcinoma of the prostate. The tissues were fixed in Helly's fluid. Some of the sections were stained with hematoxylin and eosin; others, with the eosin-azure combination of Giemsa.

The patient died within two hours after admission to the University Hospital. Unfortunately no blood smears were secured. The clinical history is meager. The prostatic carcinoma had metastasized widely in lymph nodes, bone marrow, lungs, liver and kidneys. There were no tumor cells in the spleen. The mesenteric lymph nodes were greatly enlarged and almost completely replaced by tumor cells; in some nodes hardly a trace of lymphoid tissue persisted. The bone marrow showed extensive invasion. The tissues of primary interest in this connection were the liver, the spleen and the bone marrow.

### CLINICAL HISTORY

The patient was admitted to the University Hospital at 5 p. m., complaining of shortness of breath and swelling of the feet and ankles of about ten days' duration. He had been in good health up until six or seven months prior to this, at which time he began to suffer with pain in the back and legs. He had no swelling of his feet and legs at that time. During the past two weeks he had had a troublesome cough that was productive of a purulent sputum. He had lost "a lot of weight" during the past year.

Physical examination revealed a well developed and well nourished white man about 63 years of age who was markedly dyspneic. The retinal vessels showed arteriosclerotic changes. Pyorrhea was present. The tonsils were chronically diseased. The respirations were rapid and shallow. There was dulness to percussion from the level of the fourth dorsal spine posteriorly to the base of the lung on the

left. The apex beat could be neither felt nor heard. The heart sounds were very distant but regular. The abdomen was distended. The edge of the liver extended about 3 fingerbreadths below the costal margin and was quite tender. The prostate was normal in size and regular in contour. There was an area of suspicious hardening in the left outer lobe. There was pitting edema of the feet and ankles and over the sacrum. On admission his temperature was 100.8 F. The pulse rate was 108. No examination of the blood was made. The patient died one hour and forty minutes after admission to the hospital. While he was in the hospital his chest was tapped and a bloody fluid obtained. Caffeine and epinephrine were given to him as emergency measures. A diagnosis of hypertensive heart disease with congestive failure was made. The bloody pleural effusion was explained as due probably to a pulmonary malignant process, possibly metastatic.

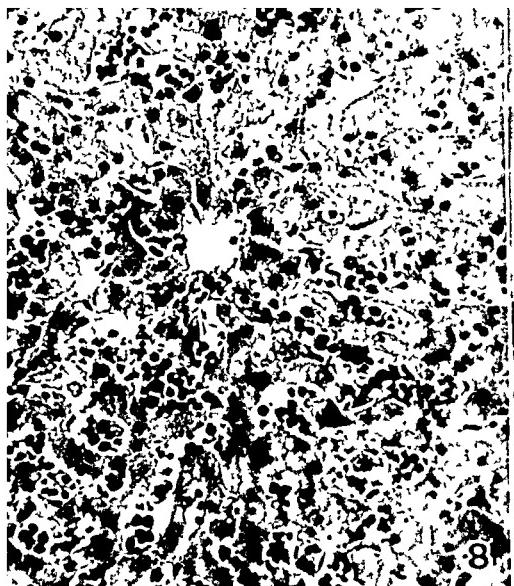
#### AUTOPSY REPORT

Since the data pertain largely to the metastasizing carcinoma, it seems unnecessary to include them here.

*Anatomic Diagnosis.*—Adenocarcinoma of the prostate, with metastases to pelvic, retroperitoneal, abdominal, axillary and inguinal lymph glands and to the liver, lungs, peritoneum, pleurae, adventitial layer of the pulmonary artery and bone marrow; extensive extramedullary formation of blood in the liver, spleen and peri-renal fat; serosanguineous pleural and pericardial effusions; thrombosis of small branches of the pulmonary arteries; infarction of the spleen; fibrosis of the pancreas with hypertrophy of the islands of Langerhans; generalized arteriosclerosis; scarring of the heart muscle and kidneys; Mönckeberg's sclerosis of peripheral arteries; slight cardiac hypertrophy (left ventricle); nephrolithiasis with hemorrhage into the renal pelvis; chronic cholelithiasis and appendicitis; abdominal adhesions; atrophy of the testes.

#### HEMOPOIESIS

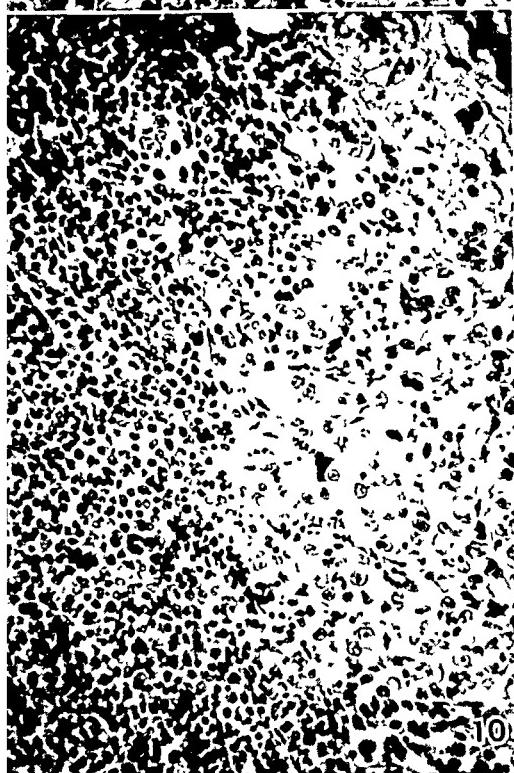
*Liver.*—The striking feature of the liver was the condition of the intralobular capilliform sinusoids. Formation of red cells was very active within the sinusoids. In many regions the sinusoids were almost completely filled with collections of cells including all stages of erythropoiesis (fig. 8). The most conspicuous cell was the relatively large hemocytoblast, with a large, vesicular, finely granular nucleus, one or several nucleoli and basophilic cytoplasm (figs. 9 and 22). These cells occurred in large numbers; they appeared identical with those of similar groups in the bone marrow and spleen; many were in mitosis. No evidence of a local origin appeared; the reticulum cells and endothelium were hemopoietically inactive. The Kupffer cells of the sinuses were phagocytically active; many of the cells contained ingested erythroblasts. The source of origin of these groups of hemocytoblasts and maturing cells within the hepatic sinusoids was obviously the spleen; the hemocytoblasts were swept to the liver via the splenic vein-portal vein route, and continued maturation under the favorable erythropoietic condition of the relatively stagnant venous blood of the sinusoids.



8



9



10



11

#### EXPLANATION OF PLATE 2 (CASE 2, ADENOCARCINOMA OF PROSTATE)

Fig. 8.—Section of a hepatic lobule. The central vein is empty. The capilliform sinusoids contain groups of hemocytoblasts and maturing erythrocytes. Helly fixation; hematoxylin and eosin stain;  $\times 180$ .

Fig. 9.—Area of hepatic tissue showing a capilliform sinusoid filled with blood cells, including one hemocytoblast (indicated by arrow) and a number of erythroblasts and normoblasts;  $\times 800$ .

Fig. 10.—Area of femoral marrow, showing large mass of tumor cells at the right;  $\times 180$ . The tumor-free marrow is predominantly granulocytopoietic.

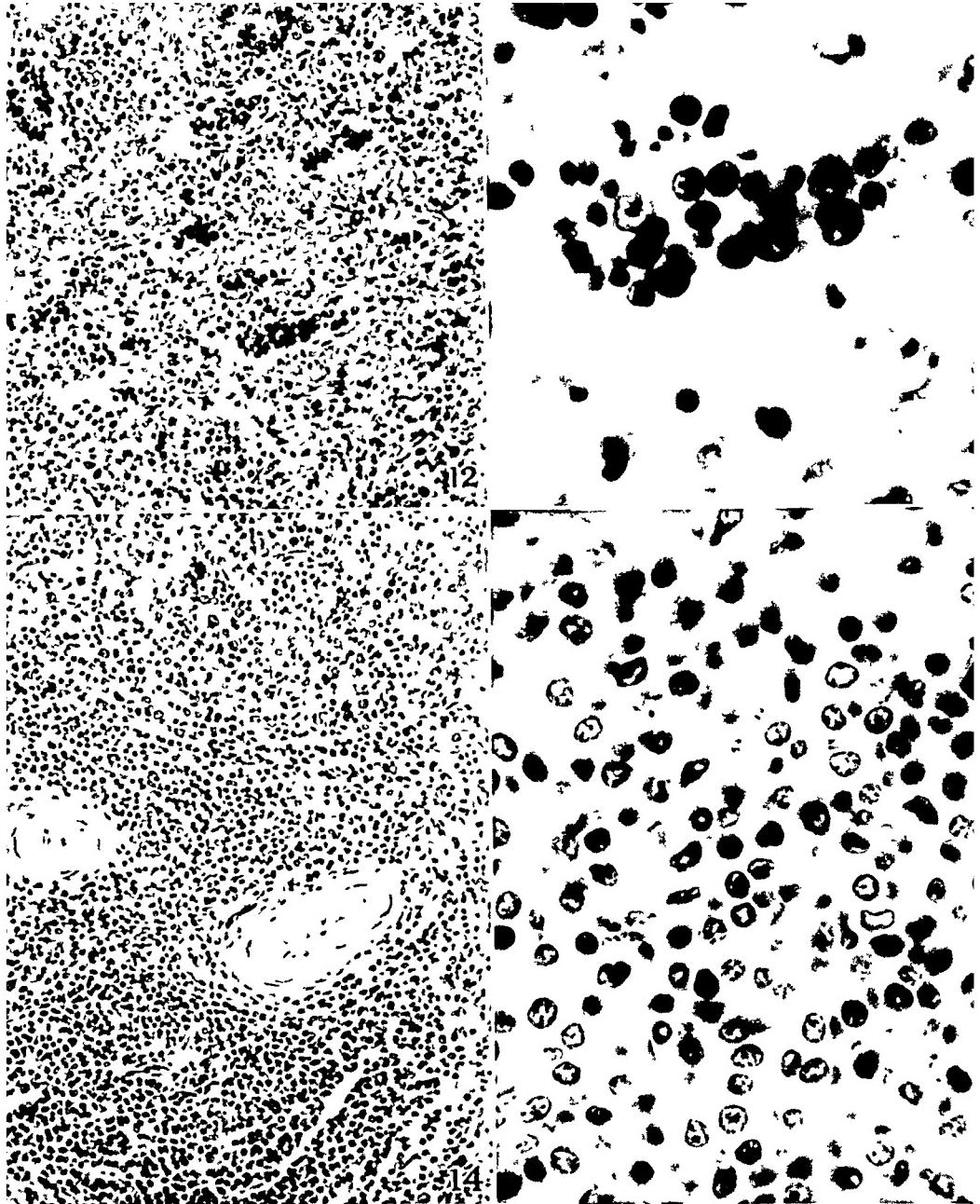
Fig. 11.—Relatively normal area of the femoral marrow of the section reproduced in figure 10, showing two groups of maturing erythrocytes, including a few hemocytoblasts;  $\times 800$ .

In certain regions the periportal area also showed hemocytopoietic tissue. The cells were mostly of the small lymphocyte type. These were apparently of local origin. The areas contained typical large hemocytoblasts, normoblasts and many cells of intermediate stages between small lymphocytes and hemocytoblasts.

*Bone Marrow.*—The erythrocytopoietic tissue of the bone marrow was greatly restricted through invasion by the tumor (fig. 10). Only femoral marrow was removed at autopsy. However, the specimen had a length of approximately 6 cm.; all of this was extensively invaded by tumor cells. In view of the conditions in the femur it seems legitimate to assume that bone metastasis was widespread. In a typical section of the femoral marrow there remained centrally a core of apparently normal tissue. Both granulocytopoiesis and erythrocytopoiesis were active. The groups of hemocytoblasts and maturing red cells here were identical with those in the hepatic sinusoids (fig. 20). In general, however, the groups were smaller. These groups occurred in sinuses, distinct from the granulocytopoietic areas (fig. 11). Peripherally, in those regions free from metastasis, hemopoiesis was almost exclusively of the granular type. The appearance of the tumor-free marrow as a whole was one of very active granulocytopoiesis and meager erythrocytogenesis. There was no evidence of local endothelial derivation of stem cells.

*Spleen.*—The spleen was more than twice its normal size. It measured 17 by 10.5 by 5 cm., and weighed 510 Gm. It was extremely active in the formation of red cells. It contained no tumor cells. Eosinophilic and neutrophilic granulocytes were rare and uniformly of definitive type; they were obviously not formed locally. In other respects the spleen looked like bone marrow (fig. 12) except for the small lymphoid (splenic) nodules (fig. 14); megakaryocytes were almost as numerous as in the femoral marrow. Basophils occurred in small numbers and were of local origin; they included all developmental stages from hemocytoblasts to definitive mast cells.

Primary interest concerns the origin of the very numerous hemocytoblasts, from which the red cells arose. The hemocytoblasts occurred in groups, with mingled maturing erythrocytes as in the liver and the bone marrow (figs. 13, 17, 18 and 19). Concerning the origin of the hemocytoblasts two possibilities are presented: (1) they may have been carried here from the marrow; (2) they may have arisen from the splenic parenchyma. The first possibility seems contradicted by the fact that the spleen contained very few granulocytes. If the hemocytoblasts and maturing erythrocytes of the splenic sinuses had been brought here from the marrow, it would be expected that granulocytes also were included, for they are abundant in the marrow. Accordingly, the hemo-



EXPLANATION OF PLATE 3 (CASE 2, ADENOCARCINOMA OF PROSTATE)

Fig. 12.—Area of splenic pulp;  $\times 180$ . The sinuses are well filled with groups of hemocytoblasts and maturing red cells.

Fig. 13.—Splenic sinus (lowermost sinus in figure 12) showing a large group of blood cells; including hemocytoblasts, erythroblasts and normoblasts;  $\times 800$ .

Fig. 14.—Splenic nodule, showing two arterioles;  $\times 180$ . The area of transition between the nodule and the splenic pulp appears about midway between the arterioles and the upper edge of the figure. The predominating cell of the nodule is the small lymphocyte.

Fig. 15.—Area from the splenic nodule of figure 14;  $\times 800$ . The periphery of the nodule shows in the upper portion of the figure. Small lymphocytes predominate. Cells transitional between small lymphocytes and hemocytoblasts are numerous. These nodules consist of cells in two stages of the lymphoid hemocytoblast: hemocytoblast and lymphocyte.

cytoblasts of the spleen presumably represented differentiation products of the local lymphocytes, derivatives of local reticular cells. There was no evidence that the local reticular or endothelial tissue functioned directly as a source of origin of the hemocytoblasts. It appears, then, that the spleen had reverted to a fetal condition as an erythrocytopoietic organ in compensation for the restriction of the normal erythrocytopoietic tissue in the marrow. Since the lymph nodes were almost entirely replaced by tumor tissue, the spleen must have compensated also in the production of additional lymphocytes. This obligatory, compensatory activity on the part of the spleen for both the lymph nodes and the red marrow presumably explains its great hypertrophy.

*Lymph Nodes.*—The majority of the lymph nodes, especially those of the mesenteric group, were greatly enlarged and except for an occasional trace of lymphoid tissue consisted of tumor cells; the appearance closely resembled that of the prostate.

In those lymph nodes in which the parenchyma had been only in part replaced by the tumor, the remaining lymphoid tissue was greatly altered; it closely resembled the spleen. The cortical nodules were confluent and only vaguely outlined; germinal centers were lacking. The sinuses were large and contained considerable blood. The predominating cell of the cortical region was the small lymphocyte. The medullary cords contained small groups of hemocytoblasts and adjacent maturing erythrocytes. There occurred also, especially along the corticomedullary boundary, large numbers of cells in transitional stages between small lymphocytes and hemocytoblasts. The erythrocytopoietic areas contained also numerous megakaryocytes. The tumor-free portion of the lymph nodes was converted into hemolymph gland tissue, the red cells arising from hemocytoblast derivatives of small lymphocytes.

Certain small lymph nodes as well as certain portions of the larger lymph nodes, both tumorous and tumor-free, contained considerable fat. Such tissue closely resembled red marrow. Formation of red cells was active, and megakaryocytes were abundant.

Most striking were the smaller tumor-free lymph nodes. These were genuine hemolymph nodes. There was no sharp demarcation between the cortex and the medulla. However, the cortical area showed a predominance of small lymphocytes. Certain slightly more compact regions probably marked the site of the original cortical nodules. The sinuses were well filled with blood. They contained also large numbers of hemocytoblasts and maturing erythrocytes. The parenchyma also contained hemocytoblasts and normoblasts as well as many cells in transitional stages between small lymphocytes and hemocytoblasts. Both sinuses and parenchyma contained also many megakaryocytes. No evidence appeared of phagocytic activity on the part of these cells. They

represented hypertrophied hemocytoblasts. The evidence from this material supports my earlier conclusion regarding the significance of hemolymph nodes (Jordan<sup>7</sup>): The evidence favors the interpretation in terms of a modified lymph node in which the small lymphocytes give rise to hemocytoblasts which differentiate into erythrocytes.

*Fat Tissue.*—The perirenal fat in the region of the hilus contained, adjacent to the renal papillae, numerous small well vascularized patches of erythrocytopoietic tissue. The predominating cell was the small lymphocyte. However, groups of hemocytoblasts and cells in all stages of the maturation into erythrocytes also occurred here. Some of these patches contained also, especially numerously peripherally, macrophages filled with yellowish-brown globules of variable size, presumably erythrocyte débris. It is possible that this hemocytopoietic tissue, like marrow, became active in destruction as well as in formation of red cells. Some of the patches contained also numerous megakaryocytes. Similar hemocytopoietic patches occurred in the perinodal fat in the sections of the lymph nodes and in the peripancreatic fat. The histologic conditions resembled closely those recently described by Blaisdell<sup>8</sup> in a retroperitoneal lipomatous tumor.

#### ERYTHROID METAPLASIA OF THE SPLEEN IN CASE 2

The genetic history of the splenic erythrocytes began with the lymphocytes of the nodules (malpighian follicles). The splenic nodules were small. They consisted almost exclusively of typical small lymphocytes (figs. 15 and 16). Peripherally there were sometimes considerable numbers of typical hemocytoblasts (fig. 19). Many of these were in mitosis. In the intermediate regions of the splenic nodules there occurred transitional stages between small lymphocytes and large hemocytoblasts. The splenic nodules lacked germinal centers (secondary nodules). This condition suggests the approach to a state of functional exhaustion. There was no proliferation of the small lymphocytes. However, hemocytoblasts both of the nodules and of the sinuses multiplied actively by mitosis.

The hemocytoblasts which originated from lymphocyte ancestors in the splenic nodules were carried into the splenic sinuses. Here they underwent maturation into red blood corpuscles (figs. 17 and 18). A certain number hypertrophied (fig. 21) and became ancestors of megakaryocytes. The megakaryocyte in later stages contained an extensively lobulated nucleus. There was no evidence that it arose as a fusion product of hemocytoblasts; the cell was mononucleated, the complex condition of the definitive nucleus representing a modified originally spheroid body. Nor was there any evidence that the splenic giant cell was capable of phagocytosis; it presumably had the same function here as in the bone marrow.

The dominating cell of the spleen was the hemocytoblast. In typical form it was a spheroid cell with a relatively large nucleus. It had a diameter of about 12 microns. In sections stained with eosin-azure the cytoplasm took a deep lilac color. The nucleus stained only lightly; it appeared vesicular; the delicate

7. Jordan, H. E.: Am. J. Anat. **38**:255, 1926.

8. Blaisdell, J. L.: Arch. Path. **16**:643, 1933.

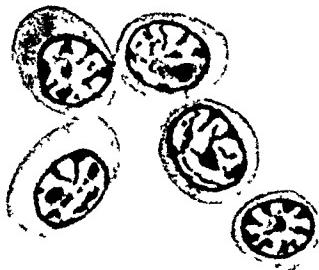
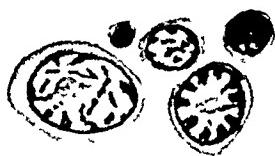
reticulum was sparse, forming a wide-meshed net. The most conspicuous feature of the nucleus was the plasmosome (nucleolus). This body stained lightly in contrast with portions of the more chromatic nuclear reticulum. It was commonly of spheroid shape; but it sometimes was bilobed, or even of triangular or rectangular form in sections, with the edges continuous with the more deeply staining reticulum. Many hemocytoblasts contained two or three plasmosomes.

In the splenic nodules, as stated, occurred numerous cells that were transitional between the central typical small lymphocytes and the peripheral typical hemocytoblasts. The typical small lymphocyte had a diameter of about 8 microns. It had a spheric shape. It had relatively little cytoplasm; this stained light blue or lilac with eosin-azure. The spheric nucleus contained a coarse reticulum; the threads ended on the nuclear membrane in an expansion. This gave an appearance of peripheral arrangement of angular chromatin blocks. The reticulum frequently attached centrally to a larger angular chromatin mass. In lightly stained preparations this central chromatin nucleolus was seen to consist mainly of a spheric plasmosome. In the intermediate stages between the small lymphocyte and the hemocytoblast the cell attained an intermediate size, the nucleus became more vesicular, the nuclear reticulum became finer and more widely meshed, and the nucleolus lost its chromatin and appeared as a plastin body; meanwhile the cytoplasm changed from light blue or lilac to deeper lilac color. Without prejudice as to the nature of the small cell of the splenic nodule, whether a small lymphocyte or a small hemocytoblast, the evidence seems conclusive that this small cell was the ancestor of the larger typical hemocytoblast which changed into a normoblast within the splenic sinuses.

The hemocytoblast in the spleen underwent the well known changes characteristic of erythropoiesis (figs. 17 and 18). These changes were identical in the splenic sinuses, marrow sinuses (fig. 20) and hepatic sinuses (fig. 22). Both cytoplasm and nucleus became condensed. The chromatin blocks became arranged peripherally, giving the nucleus the cart-wheel character typical of the erythroblast. Meanwhile, in eosin-azure preparations, the lilac color of the cytoplasm changed to pink or red. The process of nuclear and cytoplasmic condensation continued until the typical normoblastic stage with a dense chromatic nucleus was attained. At this stage, more frequently in the case of the normoblast of the hepatic sinuses, loss of nucleus in the formation of the erythroplastid was preceded by karyorrhexis; in this condition the nucleus frequently assumed a trefoil or multiglobular shape.

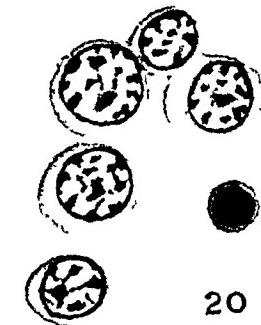
#### COMMENT

This case illustrates the reversal of the phylogenetic history of the blood-forming tissues. In phylogeny the fundamental blood-forming organ, the spleen, apportions its functions of lymphocytopoiesis and erythrocytopoiesis at the higher levels, respectively, among lymph nodes and bone marrow. It retains prominently in the mammalian adult only the functions of lymphocyte and monocyte formation. Since the lymph nodes also perform these functions to a high degree, the spleen represents as regards its primary function of blood formation only a vestigial organ. However, by virtue of its reticular stroma, its lymphocyte parenchyma and its sinusoidal venous circulation, it retains its evolutionary and fetal potentiality for the formation of the red cells. In



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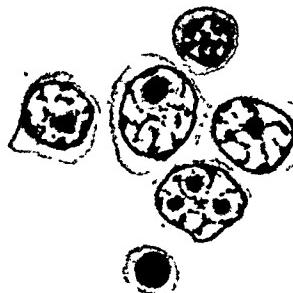
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**EXPLANATION OF PLATE 4 (CASE 2, ADENOCARCINOMA OF PROSTATE)**

Fig. 16.—Group of six lymphocytes from a splenic nodule. The two larger lymphocytes (above at the right) are approaching the size and features of the hemocytoblast. Helly fixation; hematoxylin and eosin stain;  $\times 1,350$ .

Fig. 17.—Group of six cells from a splenic sinus. The two larger cells are typical hemocytoblasts. The smaller cells with pyknotic nuclei are normoblasts. The two cells of intermediate size above at the right are erythroblasts. Helly fixation; hematoxylin and eosin stain;  $\times 1,350$ .

Fig. 18.—Similar group of cells from a splenic sinus.

Fig. 19.—Group of five cells from a splenic sinus, including four typical hemocytoblasts and one erythroblast (below at right). Helly fixation; eosin-azure stain;  $\times 1,350$ .

Fig. 20.—Group of six cells from a marrow sinus, including two typical hemocytoblasts (at left), three erythroblasts and one normoblast (below at right). Helly fixation; hematoxylin and eosin stain;  $\times 1,350$ .

Fig. 21.—Large hemocytoblast, ancestor of the megakaryocyte;  $\times 1,350$ .

Fig. 22.—Group of six cells from a hepatic sinus, including three hemocytoblasts, one erythroblast (above) and one normoblast. Helly fixation; hematoxylin and eosin stain;  $\times 1,350$ .

this case of adenocarcinoma of the prostate both the lymph nodes and the bone marrow were largely eliminated from the hemocytopoietic system by reason of extensive metastases, and the spleen was stimulated to assume as a compensatory measure its original erythrocytopoietic activity. The condition roughly parallels the evolutionary level of the Amphibia in which the bone marrow has only slight erythropoietic activity, the spleen being the dominant organ in the production of red cells. From another point of view this case represents a natural experiment in which most of the lymph nodes and large portions of the bone marrow have been eliminated from the hemopoietic system. The resulting condition provides the stimulus for the compensatory hyperfunction of the remaining potentially erythrocytopoietic tissue, namely, the spleen.

The interpretation of the small lymphocyte of the splenic follicles as the ancestor of the erythrocytogenic hemocytoblast is based on two sets of data: (1) transitional stages; (2) the phylogeny of hemocytopoietic tissues. The evidence from transitional stages must be evaluated in connection with the spatial relationships and in comparison with conditions in the lymph nodules of normal active lymph nodes and the normal spleen.

As described in a foregoing paragraph, the nodules of the erythrocytopoietic spleen in case 2 lacked germinal centers. The nodules consisted almost exclusively of typical small lymphocytes. Along the periphery occurred a number of typical hemocytoblasts; in the region between the central and the peripheral area occurred numerous cells that were transitional between the small lymphocyte and the hemocytoblast with respect to both size and structure. In the extranodular regions, especially numerous in the venous sinuses, occurred large numbers of hemocytoblasts mingled with cells in all stages of maturation into erythrocytes. This evidence, considered in relation to the fact that many of the nodules had disappeared and that those which persisted were relatively small, suggests the conclusion that the nodules were being used up in the production of hemocytoblasts by metamorphosis of the small lymphocytes.

Considering now the nodules of normal lymph nodes and the normal spleen, the following facts emerge. These nodules contain large germinal centers, composed chiefly of large cells with lightly staining nuclei cytologically identical with hemocytoblasts. These cells, here designated as lymphoblasts, represent free derivatives of the reticular stroma of this region. While the cells are generally of large size, taken as a group they include small, medium-sized and large varieties. They are collectively characterized by a vesicular nucleus, with delicate reticulum and one or several nucleoli. The small varieties may grow to larger size. Only the cells of maximum size divide; the stages in mitosis are numerous. Peripheral to the germinal center occurs a compact layer of

variable width consisting of concentric rows of small lymphocytes. These represent daughter cells of the hemocytoblasts. As typical small lymphocytes they find their way into the medullary cords and sinuses and eventually into the blood stream. Meanwhile a certain number enlarge to become large lymphocytes cytologically identical with the original lymphoblasts (hemocytoblasts). In view of this history it becomes meaningless to describe a small lymphocyte in terms of age; it is both the offspring of a hemocytoblast and the parent of one. The small lymphocyte of the lymph node, splenic parenchyma and peripheral blood is a relatively undifferentiated cell capable of the expression of any one of its multiple developmental potentialities depending on environmental stimuli. Thus it may be ancestral to an erythrocyte, a granulocyte or a monocyte. Accordingly, it is misleading to speak of the small lymphocyte of the blood as a definitive cell comparable to the erythrocyte. Both nuclear and cytoplasmic features mark it as a relatively undifferentiated cell. It represents in fact a hemocytoblast reduced in size presumably for purposes of more ready transportation. The smaller forms may enlarge, thus contributing the numerous medium-sized ones.

The foregoing conclusion is not invalidated by the fact of the occurrence of degenerating and senile small lymphocytes in the lymph nodes, in the spleen and in the peripheral blood. Degenerate forms are especially numerous in areas of very active production. Hu and Ch'in<sup>9</sup> reported that 21.2 per cent of the small lymphocytes in the inguinal lymph nodes of the rat were degenerate, and 28.9 per cent of those in the spleen. The abortive result represents a casualty of rapid formation. Young cells as well as old cells may be defective or injured and as a result degenerate. Similarly, in the blood stream a certain small number may degenerate; these are the senile or "definitive" type recognized by Hu and Ch'in<sup>8</sup> and Wiseman.<sup>10</sup> Hu and Ch'in gave the percentage in rat's blood as 0.15. The striking and significant fact in the work of Hu and Ch'in with supravital stained preparations relates to the small number of so-called definitive forms, and the large number of intermediate forms.

According to Wiseman,<sup>10</sup> 45 per cent of the small lymphocytes of the peripheral blood of the rabbit are of "mature" type, judged by the relatively lesser degree of cytoplasmic basophilia and the decreased content of the mitochondria in supravital preparations of such cells as compared with the younger forms. About 5 per cent are recognized as in the "end phase," lacking all of the characteristics of youth and maturity. There can be no question regarding the degeneration of a certain small

9. Hu, C. H., and Ch'in, K. Y.: Proc. Soc. Exper. Biol. & Med. **30**:433, 1933.

10. Wiseman, B. K.: J. Exper. Med. **54**:271, 1931; Folia haemat. **46**:346, 1932.

percentage of circulating lymphocytes. However, the so-called mature small lymphocytes may still have the youthful character of possessing metachromatic granules as revealed in Wright's stain. And cytoplasmic basophilia is not necessarily indicative of youth; the cytoplasm of the reticular cell ancestor of the lymphocyte is less basophilic than that of the free young lymphocyte, while the cytoplasm of the plasma cell is more basophilic than that of the ancestral lymphocyte. The best single criterion of the age of the lymphocyte is the character of the nucleus. The degenerating lymphocyte contains a relatively small, deeply pyknotic nucleus.

Approached from the standpoint of the evolutionary history of the blood-forming tissues the matter appears thus: The spleen is the fundamental erythrocytogenic organ from lower fishes through amphibians. In reptiles and birds and even in certain mammals, e. g., the bat, the hedgehog, the opossum, it maintains a considerable degree of activity as a site of red cell production. Also in fetal life in man it functions largely as an erythrocytopoietic tissue. Only when marrow appears in phylogeny at the level of the amphibians, as an incident of hollow bones, in human ontogeny during the second month, is the production of red cells partly shifted to this tissue. At the level of the mammals the originally multiple function of the spleen becomes apportioned to the lymph nodes and the bone marrow; as regards the original specific function of forming blood the spleen of the mammal represents a vestigial organ.

The erythrocytopoietic lymph nodes in case 1 contrast sharply with those in case 2. Whether the difference signifies only separate phases of the same process or something more fundamental remains uncertain. Both were active in the formation of red cells, and the normal demarcation between the cortex and the medulla was largely obliterated; but otherwise the nodes were very dissimilar. In case 1 (leukemia) the parenchyma was uniformly moderately dense; the sinuses were not especially prominent and contained relatively few erythrocytes. Megakaryocytes were very rare, and macrophages were not numerous. Maturing erythrocytes were uniformly scattered among the lymphocytes of the parenchyma. This lack of segregation of maturing erythrocytes in small groups was perhaps the most characteristic feature of these lymph nodes. Typical hemocytoblasts, such as occurred in large numbers in case 2, were relatively rare. Those that occurred, however, appeared in small groups. These lymph nodes did not closely resemble typical hemolymph nodes. The large number of cells that were transitional between typical small lymphocytes and erythroblasts suggests that in these nodes the erythrocytes differentiated directly from small and medium-sized polyvalent lymphocytes instead of from hemocytoblasts as in case 2.

The tumor-free "lymph nodes" in case 2 (adenocarcinoma of the prostate) were typical hemolymph nodes. The sinuses were well filled with blood; megakaryocytes were numerous; cells transitional between small lymphocytes and hemocytoblasts and between the latter and erythroblasts were abundant. Both hemocytoblasts and maturing erythrocytes were seen occurring in groups. On the basis of the information from an earlier comparative study of hemolymph nodes in the sheep, rabbit and dog and man (Jordan<sup>11</sup>) the conclusion is suggested that the nodes in case 2 signify later stages of the process of erythrocytopoietic metaplasia represented in case 1.

The evidence from these two cases of extramedullary formation of blood in man emphasizes the relative independence of the two chief aspects of marrow hemopoiesis: erythrocytopoiesis and granulocytopoiesis. Here the two processes are separated as at the lower evolutionary levels and in the mammalian fetus during the period when the spleen is active in the production of red cells. The very intimate association of the two processes in the bone marrow of adults obscures their essential independence. However, both erythrocytes and granulocytes arise from apparently identical ancestors, the lymphoid hemoblasts. But the differentiation of granulocytes occurs extravascularly; the production of erythrocytes, generally only intravascularly. The differential factors leading respectively to red cell or to granulocyte formation are apparently entirely environmental.

In the primitive spleen of the cyclostome fishes erythrocytopoiesis and granulocytopoiesis are associated; their spleen represents a myeloid tissue equivalent to the bone marrow of mammals. In the lung fishes the spleen still includes both tissues, but granulocytopoiesis is also very active in the wall of the intestine (Jordan and Speidel<sup>12</sup>). In elasmobranch fishes granulocytopoiesis is restricted to the stroma of the gonads and formation of red cells occurs only in the spleen. In most teleost fishes red cells are formed in the spleen and granulocytes in the mesonephron; in some teleosts both processes occur to a variable degree in the kidney. In most salamanders the formation of red cells is restricted to the spleen and the formation of granulocytes to the subcapsular stroma of the liver; in a few salamanders, e. g., *Proteus*, the differentiation of granulocytes is restricted to the kidney. In frogs, during the greater part of the year, the red cells arise in the spleen and the granulocytes to a large extent in the marrow; however, for a brief period after hibernation both processes occur in the bone marrow. At all of these levels the maturation of the red cells occurs to a variable extent in the periph-

11. Jordan, H. E.: *J. Morphol. & Physiol.* **44**:89, 1927.

12. Jordan, H. E., and Speidel, C. C.: *Am. J. Anat.* **46**:355, 1930; *J. Morphol. & Physiol.* **51**:319, 1931.

eral blood stream (Jordan<sup>13</sup>). Furthermore, at all levels there may be considerable granulocytopoietic activity in the wall of the intestine. In view of these facts it is in accord with a more precise terminology to speak of erythrocytopoietic metaplasia and granulocytopoietic metaplasia rather than myeloid metaplasia. The metaplasia of the lymph node and spleen in cases 1 and 2 was of the erythrocytopoietic type.

Perhaps the most significant feature of case 1 was the condition of the bone marrow; the bulk was lymphoid. The marrow had the appearance largely of lymphoid tissue; the lymph nodes and portions of the spleen had the appearance of red bone marrow; the lymph nodes had undergone an erythrocytopoietic metaplasia, the marrow a lymphoid metaplasia. The dominating cell of the bone marrow was a typical small lymphocyte. The lymphocytes appeared to be more closely aggregated about arterioles, suggesting lymph nodules. Hemocytoblasts and erythroblasts, while numerous in certain regions, occurred in relatively small numbers. In view of the large percentage of small lymphocytes in the blood the conclusion might be drawn that the lymphocytes of the marrow represented cells filtered out of the blood stream. If such a conclusion were justified it would support the idea that the normal fate of the lymphocyte is largely to serve as an ancestor of the red cell in the venous sinuses of the bone marrow. This is in part the fate of the lymphocyte of anuran amphibians (Jordan<sup>14</sup>). To account for the accumulation and persistence of lymphocytes in this marrow it is necessary to assume that the pathologic condition rendered impossible the differentiation at normal tempo of the hemogenic lymphocytes into erythrocytes. However, the possibility that these marrow lymphocytes represented cells of local origin cannot be ignored, especially in view of the fact that the lymphocytes of the lymph nodes and spleen were largely converted into red cells locally. However interpreted, the evidence suggests a very intimate relationship between erythrocytopoietic and lymphoid tissues. Here apparently, as in lower forms in which erythrocytopoiesis is restricted to the spleen, the lymphoid and erythroid tissues were potentially one and the same tissue.

#### SUMMARY

Two cases of extramedullary hemocytopoiesis with a very different fundamental pathology are described. In case 1 (acute leukemia) erythrocytopoietic metaplasia was pronounced in the lymph nodes. In case 2 (adenocarcinoma of the prostate) in which many of the lymph nodes and large portions of the marrow had been obliterated by metastases, the spleen was the seat of erythrocytopoietic metaplasia. These

13. Jordan, H. E.: Quart. Rev. Biol. **8**:58, 1933.

14. Jordan, H. E.: Am. J. Anat. **25**:437, 1919.

two cases in a sense complement each other. They represent natural experiments in which in one case the bone marrow and the spleen have been largely removed from the hemopoietic system, leaving the lymph nodes for compensatory erythrocytopoietic function; in the other case the marrow and the lymph nodes have been replaced, leaving the spleen unimpaired. In both cases the small lymphocyte functions as the ancestral cell of the erythrocyte.

In case 1 (leukemia) the lymph nodes especially, to some degree the spleen, had undergone erythroid metaplasia; the bone marrow showed extensive lymphoid metaplasia. The evidence emphasizes the intimate relationship between myeloid and lymphoid tissue with respect to erythrocytogenesis. The ancestor of the red cell was in both tissues a small lymphocyte, the lymphoid hemoblast. Such a condition parallels that of earlier ontogenetic and lower phylogenetic stages in which the lymphoid spleen is the locus of red cell formation.

In case 2 (adenocarcinoma of the prostate) the lymph nodes had been largely replaced by tumor tissue and the bone marrow was greatly restricted; the spleen had undergone a high degree of erythroid metaplasia, the formation of red cells being very active. Here the small lymphocyte first became a typical hemocytoblast, which then matured into an erythrocyte. The few small persistent lymph nodes had become converted into typical hemolymph nodes, with much blood in the sinuses, large numbers of megakaryocytes and very active transformation of small lymphocytes into erythrocytes. The evidence emphasizes the independence of erythrocytopoiesis with respect to granulocytopoiesis. The "myeloid" metaplasia of the spleen and lymph nodes concerned only the production of red cells.

The evidence supports the claim of the erythrocytogenic capacity of the small lymphocyte and the interpretation of this cell as a relatively undifferentiated one with multiple developmental potentialities.

# CHRONIC CICATRIZING ENTERITIS

WITH INVOLVEMENT OF THE CECUM AND THE COLON

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AND

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The intestinal lesions varyingly described as regional ileitis, chronic cicatrizing enteritis and nonspecific granuloma of the intestine have received an increasing amount of attention since they were first described by Braun,<sup>1</sup> in 1909, on the basis of one case in which inflammation of the sigmoid flexure was treated by surgical removal and two cases in which the mass felt at operation in the cecum disappeared without resection. The description of this condition by Braun threw light on certain conditions simulating malignant changes in the intestine, which cleared up with palliative treatment, such as short circuiting and colostomy, examples of which had been reported by Moynihan<sup>2</sup> and Mayo Robson<sup>3</sup> some years previously.

Until after the war but little attention was devoted to this condition. With increased interest in malignant disease, the frequency with which this lesion simulated carcinoma brought more and more attention to it. A review of the subject was made by Tietze,<sup>4</sup> in 1920, who added a number of cases of his own, in two of which the condition was localized in the cecal region, and in all of which the lesions simulated malignant disease and were found to be examples of "non-specific granuloma."<sup>5</sup>

A second review of the infectious granulomas, not only those of the intestinal tract but also those throughout the abdominal cavity, was presented in 1931 by Mock,<sup>6</sup> who brought the subject well up to date.

The picture presented by this lesion, which is better known as chronic cicatrizing enteritis,<sup>7</sup> although varied, nevertheless has a number of fairly constant characteristics. The etiology is uncertain. A number of factors have been incriminated, from inflammatory processes in the

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From the Laboratory of Pathology of the New England Deaconess Hospital.

1. Braun, Heinrich: Deutsche Ztschr. f. Chir. **100**:1, 1909.

2. Moynihan, B.: Edinburgh M. J. **21**:228, 1907.

3. Robson, A. W. M.: Brit. M. J. **1**:425, 1908.

4. Tietze, Alexander: Ergebni. d. Chir. u. Orthop. **12**:211, 1920.

5. Moschcowitz, Eli, and Wilensky, A. O.: Am. J. M. Sc. **166**:48, 1923.

6. Mock, H. E.: Surg., Gynec. & Obst. **52**:672, 1931.

7. Harris, F. I.; Bell, G. H., and Brunn, Harold: Surg., Gynec. & Obst. **57**:637, 1933.

neighborhood of the appendix to the use of certain types of suture. In many of the reported cases the condition followed an abdominal operation.

In the more characteristic forms there is fairly close restriction to the region of the ileocecal valve, although a number of cases in which the lesion was found elsewhere in the intestinal tract, and especially in the sigmoid flexure, have been reported. Even in cases with extensive cecal involvement, the appendix may show no abnormalities.

Since the most common point of involvement is the lower part of the ileum, the name regional ileitis<sup>8</sup> has come into prominence.

The symptomatology is usually vague, with abdominal cramps and other evidences of obstruction as prominent features, although these are not constant. The presence of a mass is also fairly frequent. Thus, the most common diagnosis is carcinoma, although tuberculosis and syphilis have also been confused with this condition. Usually the patient has not lost an appreciable amount of weight, is not prostrated and makes an uneventful recovery after the obstruction is removed. In some cases the development of fecal fistulas, either postappendical or spontaneous, has been a prominent feature of the clinical course of the disease.

From the standpoint of pathology, there are a number of points in common, in spite of the variation of the detailed picture. Usually the involved portion of the intestine is firmly fixed to the adjacent structures. Often, through chronic inflammatory changes in the tissues and the development of adhesions in the neighborhood of the lesion, an appreciable mass is built up. The lymph nodes draining the region show evidence of hyperplasia.

On opening the intestine, varying degrees of thickening of the wall and of ulceration of the mucosa, with stenosis, are noted.

Röntgen examination not infrequently reveals changes simulating those in an obstructing malignant lesion. There is no definite age limit, there being a wide distribution in regard to age. There is also no predilection for either sex.

We present one case which is of interest because of the involvement of the entire cecum and all the ascending colon, without any appreciable change on the proximal side of the ileocecal valve other than a moderate degree of hypertrophy of the muscularis produced through partial obstruction at the valve.

#### REPORT OF A CASE

*History.*—A woman, 62 years of age, was admitted to the New England Deaconess Hospital on Nov. 28, 1933, under the care of Dr. Howard M. Clute, surgeon of the Lahey Clinic. Eighteen years before, she had undergone cholecys-

8. Crohn, B. B.; Ginzburg, Leon, and Oppenheimer, G. D.: J. A. M. A. 99:1323, 1932.

tostomy for the relief of pain in the right upper quadrant with jaundice. She had had occasional attacks of abdominal distress since that time, which were different from the previous pain and without jaundice. She remained fairly well until four years before examination, when she had attacks of abdominal cramps, constipation and much flatus for several months. After these attacks passed without treatment, she remained well until two and one-half months before examination, when another attack of mild abdominal cramps occurred, each lasting for from several seconds to a minute. This was accompanied by marked constipation. Diarrhea was induced by catharsis. There was no vomiting. Within this period she lost about 5 pounds (about 2.3 Kg.).

The results of the physical examination were essentially negative, with the exception of a scar in the right upper quadrant. The abdominal wall was lax and flabby. Some tenderness was found in the right lower quadrant. The apparently gas-filled bowel was readily palpable in the right lower quadrant. Pelvic and rectal examinations gave essentially negative results.

Roentgen examination on December 30 showed a barium sulphate enema filling the colon as far as the hepatic flexure. The colon was dilated and atonic, with apparently a definite lesion at the hepatic flexure. Only a thin trickle of barium passed this point. The cecum and the ascending colon were distended with air. A roentgen diagnosis of carcinoma of the hepatic flexure was made, which reenforced the clinical impression of carcinoma of the cecum. The results of laboratory examinations were essentially negative.

Operation, performed by Dr. Clute on Jan. 4, 1934, was somewhat difficult because of numerous adhesions resulting presumably from the previous cholecystostomy. A mass about the size of a lemon was found involving the cecum just above the ileocecal junction. There was no evidence of metastasis to any organ, and as the lesion was freely movable resection was done. This included about 6 inches (15 cm.) of the terminal part of the ileum, the cecum, the ascending colon and a portion of the transverse colon. The resection was of the Mikulicz type, about 4 inches (10 cm.) of ileum being sutured to the transverse colon, with the usual double-barreled colostomy. The postoperative diagnosis was carcinoma of the cecum. A moderate degree of postoperative shock was readily controlled by venoclysis and blood transfusion. The subsequent course was good.

*Pathologic Report.*—A portion of bowel 36 cm. long, with a portion of the omentum attached to the distal end and a considerable mass of adherent fat and mesentery, was examined. One mass of firm fat surrounded the proximal end. On opening the lumen of the intestine, the specimen was seen to consist of 6 cm. of terminal ileum, the cecum, the ascending colon, and a portion of the transverse colon. The ileum was dilated up to 10 cm. in circumference; its wall was thickened and hypertrophied, but there was no evidence of inflammatory change. There was a prominent ring of fibrous tissue and muscle encircling the ileocecal valve. The lumen here measured barely 0.5 cm. in diameter (fig. 1). The caput of the cecum was practically obliterated by contraction, and an irregular tunnel with ulcerated walls led into a mass of firm, injected fat at the normal site of the appendix. Section through the mass of inflammatory fat revealed this passage to be about 2 cm. long; it was fairly straight, with shaggy necrotic sides. This apparent appendical lumen ended in the midst of firm, fibrous tissue which extended throughout the mass of fat surrounding the ileocecal region.

The entire cecum and ascending colon were markedly constricted, the circumference being about 1.5 cm. Not only was there constriction, but there was also considerable shortening, as was evidenced by the plicated condition of the muscu-

laris (fig. 2). This combination of circumferential and longitudinal constriction indicated a fairly long-standing fibrotic process. The mucosa was brownish red and thin, and in places it was apparently absent, leaving bare fibrous tissue.



Fig. 1.—Lower part of the ileum, the ileocecal valve, the cecum, the ascending colon and a portion of the transverse colon. The probe lies in the lumen of the appendix.

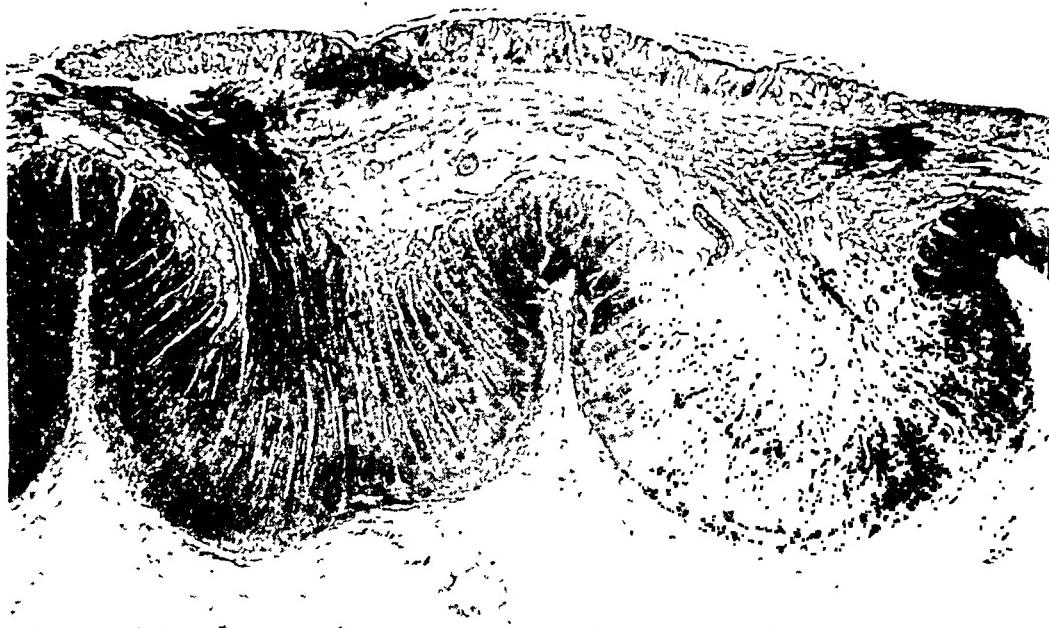


Fig. 2.—Cicatrized ascending colon just below the junction with the normal colon; reduced from a magnification of  $\times 25$ .

At the hepatic flexure there was an extraordinarily sharp and rapid change from the atrophic mucosal surface to approximately normal intestinal surface. Within a distance of about 2 cm. the colon practically regained its normal circum-

ference and the transition from the atrophic to the normal mucosa was as abrupt as though it had been cut with a knife.

*Microscopic Observations.*—Sections of the ileum were essentially normal, except for moderate hyperplasia of the musculature. The colon beyond the hepatic flexure was normal. The cecum and the ascending colon were essentially alike. The mucosa was markedly atrophic, and in numerous foci the epithelium was completely lacking. Dilated glands were scattered elsewhere, the lumens of which contained numerous polymorphonuclear leukocytes. A few of these glands in the region of the appendical orifice showed metaplasia of the epithelium, with transition to a high columnar cell with a large nucleus and little evidence of mucous secretion, suggesting the glands seen in certain carcinomas of the large intestine (fig. 3).

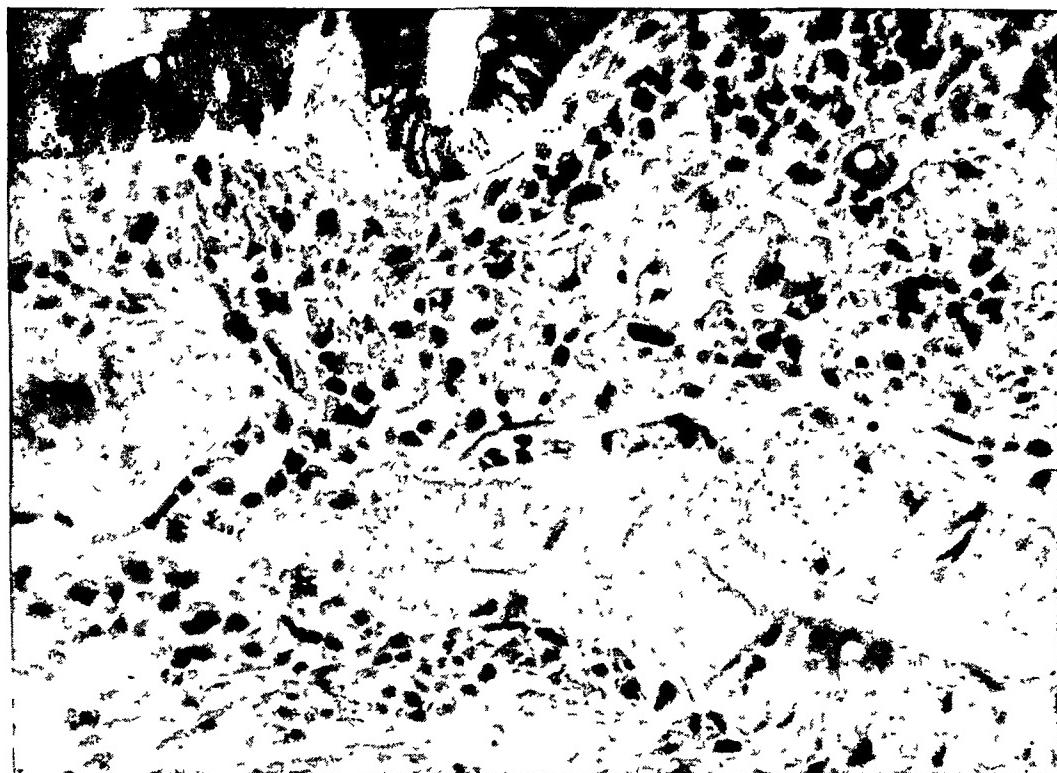


Fig. 3.—Metaplasia of the epithelium at the ileocecal valve; reduced from a magnification of  $\times 600$ .

However, no distinct evidence of invasion was found; there was no appreciable proliferation, and mitotic figures were absent. Much of the mucosa had been replaced by rather avascular granulation tissue with numerous plump fibroblasts and relatively little intercellular substance, accompanied by heavy infiltration by polymorphonuclear leukocytes, lymphocytes and plasma cells. Eosinophils were frequently encountered, particularly in the submucosa, and scattered mast cells were present. There were a few irregularly scattered foci of definite necrosis, some with a moderate amount of fibrins. Rare giant cells of the foreign body type were present in the submucosa, some of which contained vacuoles. At the point of junction of the normal colon and the involved ascending colon there was a sharp demarcation of the mucosa. A few small ulcers extended down to, or slightly below, the tunica propria, with fairly extensive extravasation of red cells beneath. The leukocytic reaction was slightly more intense here than elsewhere.

*Synopsis of Reported Cases of Chronic Cicatrizing Enteritis Involving the Cecum and the Colon*

Case	Author	Site of Lesion	Previous Operation	Appendix	Signs and Symptoms
1	Harris, Bell and Brunn <sup>7</sup> .	Portion of ascending colon, cecum, ileocecal valve and portion of ileum	Appendectomy	Absent	Acute intra-abdominal symptoms 6 months later Abdominal pain
2	Körte, W.: Arch. f. Klin. Chir. 118: 138, 1921	Portion of ascending colon, cecum, ileocecal valve and portion of ileum	None	Normal	Fistula and obstruction 3 months later
3	Wiletsky, A. O., and Moschcowitz, Eli. Am. J. M. Sc. 173: 374, 1927	Portion of ascending colon, cecum, ileocecal valve and portion of ileum	Appendectomy	Absent	Cramps in abdomen 6 years later
4	Erdmann, J. F., and Burr, G. V.: Surg., Gynec. & Obst. 57: 71, 1933	Portion of ascending colon, cecum, ileocecal valve and portion of ileum	Appendectomy	Absent	Intestinal obstruction 3 years later
5	Gordon, Donald: Ann. Surg. 97: 130, 1933	Portion of ascending colon, cecum, ileocecal valve and portion of ileum	Appendectomy	Absent	Intestinal obstruction
6	Coffen, T. H.; J. A. M. A. 85: 1603, 1925...	Portion of ascending colon, cecum and portion of ileum	Appendectomy	Absent	Intestinal obstruction
7	Jeffries, J. F.; J. M. A. South Africa 2: 184, 1928	Portion of ascending colon, cecum and portion of ileum	None	Involved	Intestinal obstruction
8	Koch <sup>8</sup> .....	Entire ascending colon, cecum and ileocecal valve	None	Not mentioned	Symptoms of gallbladder disease; colicky pain and constipation
9	Moschcowitz and Wilensky <sup>5</sup> .....	Entire ascending colon, cecum and ileocecal valve	Appendectomy	Absent	Abdominal pain and intestinal obstruction 9 months later
10	Nemilov, Alexander: Arch. f. Klin. Chir. 153: 346, 1928	Portion of ascending colon, cecum and ileocecal valve	None	Obliterated	Abdominal pain and intestinal obstruction
11	Körte, W.: Arch. f. Klin. Chir. 118: 138, 1921	Portion of ascending colon and cecum....	None	Involved	Abdominal pain
12	Moschcowitz and Wilensky <sup>5</sup> ; Erdmann's case	Portion of ascending colon and cecum....	Appendectomy	Absent	Abdominal pain and intestinal obstruction 3 months later
13	Läwen, A.: Deutsche Ztschr. f. Chir. 129: 221, 1914	Portion of ascending colon and cecum....	None	Involved	Abdominal pain
14	Läwen, A.: Deutsche Ztschr. f. Chir. 129: 221, 1914 (Law's case)	Portion of ascending colon and cecum....	None	Not mentioned	?
15	Läwen, A.: Deutsche Ztschr. f. Chir. 129: 221, 1914 (Schmidt's case)	Portion of ascending colon and cecum....	None	Normal	?
16	Golob, Meyer: M. J. & Rec. 135: 390, 1932	Cecum and ileocecal valve.....	None	Normal	Abdominal pain
17	Braun <sup>1</sup> .....	Cecum .....	None	Not mentioned	Abdominal pain
18	Braun <sup>1</sup> .....	Cecum .....	None	Not mentioned	Abdominal pain
19	Tietze <sup>4</sup> .....	Cecum with purse-string suture.....	Appendectomy	Absent	Abdominal pain
20	Tietze <sup>4</sup> ; Goto's case.....	Cecum with purse-string suture.....	Appendectomy	Not found	Intestinal obstruction 3 years later
21	Mock <sup>6</sup> .....	Cecum .....	None	Absent	Abdominal pain
22	Körte, W.: Arch. f. Klin. Chir. 118: 138, 1921	Cecum .....	Ileotomy	Normal	Abdominal pain
23	Nemilov, Alexander: Arch. f. Klin. Chir. 153: 346, 1928	Ascending colon .....	None	Obliterated	Abdominal pain 2 years later
24	Moschcowitz and Wilensky <sup>5</sup> .....	Ascending colon .....	None	Cramps and vomiting	

\* Tietze also reported four cases of typhlitis.

The muscularis was thrown into folds by longitudinal contraction of the intestine and showed extensive fibroblastic proliferation, with some edema. There were numerous fairly thick-walled blood vessels and dilated lymphatics. Polymorphonuclear leukocytes were scattered diffusely through the tissue, together with monocytes, occasional plasma cells, eosinophils and lymphocytes. Toward the serosa, mast cells were a fairly prominent feature. Large collections of lymphocytes were observed in frequent foci, with the formation of germinal centers. Giant cells of the foreign body type were seen both within, and adjacent to, these foci.

The serosa and the subserosa were markedly edematous and were thickened by a rather diffuse, loose, fibrous tissue with some evidence of fibroblastic proliferation. There were numerous large, dilated lymphatics, many of which contained polymorphonuclear leukocytes in their lumens, and in a few the Gram-Weigert stain showed gram-positive cocci in pairs and short chains. The same type of cellular infiltration occurred here as in the mucosa, with similar focal accumulation of lymphocytes. The adjacent fat was heavily fibrosed and showed infiltration by the various types of inflammatory cells already described. Minute foci of necrosis and polymorphonuclear infiltration were observed in the subserosa and the serosa.

The microscopic diagnosis was: chronic cicatrizing enteritis, with a focus of metaplasia of the epithelium; chronic appendicitis and periappendicitis, with necrosis.

#### COMMENT

We regard this case as distinct from any yet reported, because of the sharp localization of the condition to the cecum and the ascending colon and because of the remnants of a destructive, suppurative process in the vestiges of the appendix.

The sequence of events we believe to be somewhat as follows: The process began four years before the patient was examined when the attack of abdominal pain and cramps probably indicated acute appendicitis. This failed to heal properly and became the focus from which gradual extension of the inflammatory process occurred through the lymphatics and tissue spaces, with increasingly greater involvement of the large intestine. Symptoms were not marked until the low grade inflammatory process had produced a sufficient degree of constriction of the ileocecal valve and the large intestine to induce partial intestinal obstruction. This obstruction, once developed, rapidly increased in severity and led to the patient's admission to the hospital. Roentgen examination revealed the extent of the lesion, the barium enema reaching only to the hepatic flexure. What was regarded by the roentgenologist as an air-filled cecum and ascending colon was the greatly dilated, gas-filled terminal part of the ileum. The hard, inflammatory mass of fat built up about the inflamed appendix and cecum simulated malignant disease on palpation through the abdominal wall.

We did not attempt bacteriologic studies of the tissue removed, because of the obvious chronicity of the process, which would have permitted any originally causative organism to die out long before, and because the absence of mucosa over a large stretch of intestine would

permit access to the tissues of all types of organisms from the intestinal contents. It is not remarkable that focal necrosis and evidence of acute inflammation were found scattered throughout the intestinal wall, as there must have been a continual access of organisms to the unguarded tissues. While we succeeded in demonstrating streptococci with the Gram-Weigert stain within some lymphatics showing heavy polymorpho-nuclear infiltration, we do not assign an etiologic rôle to these, as there is no way of ruling them out as secondary invaders.

One point of interest is the metaplasia of the epithelium, which took place, in all probability, as a result of the long-standing abnormal condition under which the mucosa of the cecum had attempted to regenerate and maintain function. It is possible that, had this process continued longer, there might have been transformation to a malignant process. There is a strong similarity in the change seen here and that seen in the early stages of malignant degeneration of the common rectal polypus.

A study of the cases previously reported shows involvement of the cecum and the colon to be relatively infrequent. However, well established cases of this type have been reported by a number of authors; these are summarized in the table.

The cases most closely resembling ours are those of Koch<sup>9</sup> and of Moschcowitz and Wilensky.<sup>5</sup>

An important point brought out by a study of this group of cases is the high frequency of appendical involvement, emphasizing somewhat the importance of the appendical lesion in our case. Of the group, nine patients had undergone appendectomy; five showed pathologic changes in the appendix; in the case of five the appendix was not mentioned, and only four had appendixes noted as normal. In the nine cases in which the enteritis followed appendectomy, the onset of the symptoms appeared, on the average, thirteen months after the operation.

#### SUMMARY AND CONCLUSIONS

A case of chronic cicatrizing enteritis is reported, in which the condition apparently developed after appendicitis and involved the cecum, the ileocecal valve and the ascending colon.

The exact etiology is unknown. The recovery of organisms from the lesion would have been without significance, because of the ease with which the ulcerated mucosa could be traversed.

The chief importance of this lesion lies in its mimicry of carcinoma. In all probability, apparent cases of certain intestinal cancers may be explained by the fact that a lesion of this nature was mistaken for carcinoma.

9. Koch, Joseph: Arch. f. klin. Chir. 70:876, 1903.

# HEPATIC CHANGES ASSOCIATED WITH DECOMPRES- SION OF OBSTRUCTED BILIARY PASSAGES

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AND

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The recent literature contains numerous references to a group of terminal clinical syndromes vaguely designated as "liver deaths," which may occur following various operative procedures on the biliary passages. Excellent descriptions of these phenomena have been presented by Cave,<sup>1</sup> Behrend,<sup>2</sup> Stanton,<sup>3</sup> Heyd,<sup>4</sup> Connell,<sup>5</sup> Walters,<sup>6</sup> Walters and Parham,<sup>7</sup> Helwig and Schutz,<sup>8</sup> Schutz, Helwig and Kuhn,<sup>9</sup> Doran, Lewis, Denneen and Hanssen,<sup>10</sup> Bryan,<sup>11</sup> Eiss<sup>12</sup> and Weir and Walters.<sup>13</sup> The symptoms manifested by patients with these conditions have been grouped by Heyd under three headings. One group occurs in patients operated on for the relief of biliary stasis, shortly following which they slowly pass into stupor and coma, death being similar to cholemic death in unrelieved obstructive jaundice. Schutz, Helwig and Kuhn supported in general Heyd's classification but pointed out that no serious attempt has been made to investigate the morphologic changes present in the liver of these patients and that few substantial facts relative to the actual mechanisms at work in the production of

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From the pathological laboratories of the Jefferson Medical College and Hospital, the Jefferson Hospital Tumor Clinic and the Philadelphia General Hospital.

1. Cave, H. W.: Ann. Surg. **84**:371, 1926.
2. Behrend, M.: Surgical Diseases of the Gallbladder, Liver and Pancreas, Philadelphia, F. A. Davis Company, 1927.
3. Stanton, E. M.: Am. J. Surg. **8**:1026, 1930.
4. Heyd, C. G.: Am. J. Obst. & Genec. **19**:203, 1930; J. A. M. A. **97**:1847, 1931; Surg., Gynec. & Obst. **57**:407, 1933.
5. Connell, F. G.: Ann. Surg. **94**:363, 1931.
6. Walters, W.: Ann. Surg. **94**:55, 1931.
7. Walters, W., and Parham, D.: Surg., Gynec. & Obst. **35**:605, 1922.
8. Helwig, F. C., and Schutz, C. B.: Surg., Gynec. & Obst. **55**:570, 1932.
9. Schutz, C. B.; Helwig, F. C., and Kuhn, H. P.: J. A. M. A. **99**:633, 1932.
10. Doran, W. T.; Lewis, K. M.; Denneen, E. V., and Hanssen, E. C.: Ann. Surg. **98**:321, 1933.
11. Bryan, W. A.: Ann. Surg. **98**:342, 1933.
12. Eiss, S.: Ann. Surg. **98**:348, 1933.
13. Weir, J. F., and Walters, W.: J. A. M. A. **102**:93, 1934.

the clinical manifestations have been brought forward to account for them. The present study was undertaken not only to investigate these changes and to determine if possible the morphologic basis for the profound disturbances of hepatic function which occur under such circumstances, but also to observe the manner in which hepatic recuperation takes place following relief from biliary obstruction.

#### REPORT OF INVESTIGATION

The autopsy material on which this study is based was obtained from twenty cases of primary carcinoma of the head of the pancreas completely obstructing the common bile duct, in which death followed surgical decompression of the biliary passages. Five of the cases were from the pathologic laboratories of the Pennsylvania Hospital and three from the Lankenau Hospital Research Institute. Dr. John T. Bauer and Dr. Stanley P. Reimann, the respective directors, placed this material at our disposal.

The operative procedures in these cases consisted of cholecystogastrostomy, cholecystoduodenostomy, cholecystostomy or choledochostomy. Pieces of liver for histologic examination were fixed in dilute formaldehyde and Zenker's fluid; a part of the specimen was frozen and sectioned, and the remainder was blocked in paraffin, cut and stained with phosphotungstic acid, hematoxylin-eosin, scarlet red, methylene blue, van Gieson's and Mallory's connective tissue stains, iron-alum-hematoxylin stain, Wilder's<sup>13a</sup> modification of the silver diaminohydroxide stain, Mallory's potassium ferrocyanide stain and McIndoe's<sup>13b</sup> adaptation of the del Rio Hortega silver carbonate stain for biliary canaliculi. In many instances complete sets of serial sections were examined.

The gross changes in the liver, biliary ducts and gallbladder varied considerably depending on the duration of the stasis, the extent of cirrhosis, the type of operative procedure instituted for the relief of the obstruction and the interval following decompression as well as on the presence or absence of metastatic tumor nodules, abscesses, hemorrhages or degenerative hepatic lesions. It would be inappropriate to comment further on these gross changes, the possible combinations of which are so numerous and variable that no single description applies even in the majority of cases. Interest centers particularly on the microscopic changes in the liver.

*Pigmentation.*—Biliary pigmentation diminished rapidly with the disappearance of granules from the hepatic cells and of the biliary thrombi from the canaliculi and smaller ducts. Pigmentation was still discernible nineteen days after the operation, appearing as a faintly yellowish-staining material which persisted longest in cells nearest the central and sublobular veins and in those isolated by prolongations of connective tissue at the peripheries of the lobules.

*Architectural Changes.*—Disruption of the intralobular architecture and disorganization and dissociation of hepatic cell cords were constant features, being of mild degree when confined to the inner portions of the lobule (grade 1) and extensive and irregular when involving the middle and outer thirds (grade 2) or the entire lobule diffusely (grade 3). The most marked alterations occurred just beneath the capsule of the liver and in the region of the porta and larger intra-

13a. Wilder, H. C.: Am. J. Path. 8:785, 1932.

13b. McIndoe, A. H.: Arch. Path. 6:598, 1928.

hepatic biliary ducts. Both small and large groups of lobules were completely affected in twelve cases; sections from four of these showed uniform disruption of architecture throughout the entire organ (grade 4). The hepatic cells immediately bordering on portal radicles tended to maintain their normal arrangement longest, whereas the remainder were scattered about, lying singly in groups of from three to six or as short isolated cords partially supported by a meshwork of thickened reticular fibers (figs. 1 and 2).

*Regressive Changes.*—In the majority of cases some degree of regressive change was present in the hepatic cells, the entire parenchyma being involved in some instances (figs. 1 and 2). These changes were characterized chiefly by karyolysis, pyknosis and cytoplasmic disintegration. Necrosis was a constant feature in the inner portions of the lobules (grades 1 and 2), isolated cells being irregularly

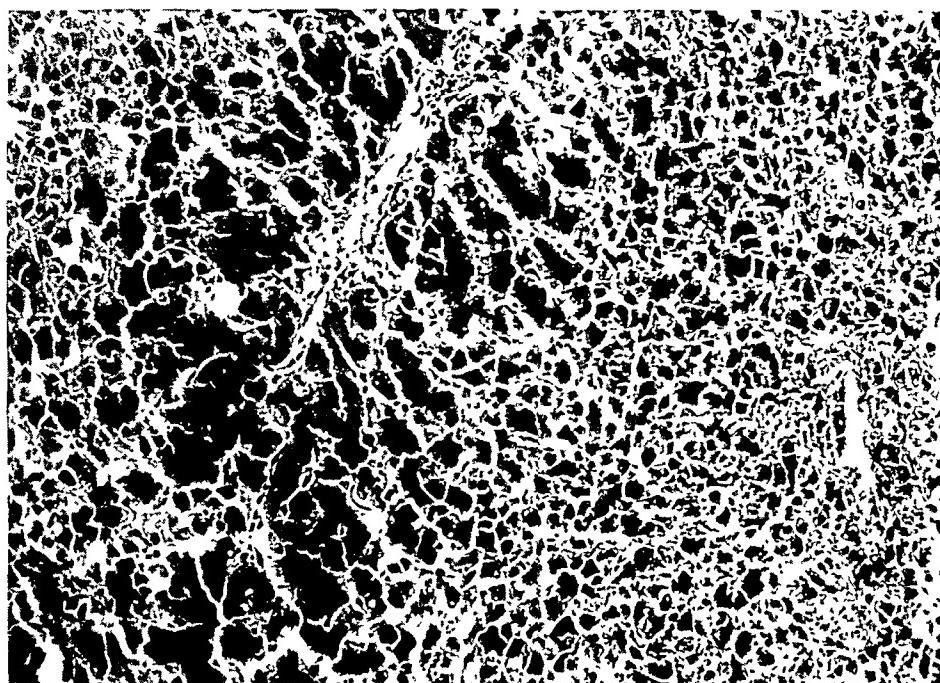


Fig. 1.—Section of the liver. A central vein lies to the right and a portal trinity in the upper left portion of the field. Note the regressive changes in the cells with disorganization and disruption of the hepatic cell cords. Reduced from  $\times 250$ .

affected in the middle and outer thirds (grades 3 and 4). Certain other cells appeared merely atrophied and distorted, alternating with necrotic cells in the two latter situations. The viable hepatic cells present in areas where disruption and disorganization were particularly well marked varied considerably in size and shape, often presenting bizarre forms resembling lymphocytes and plasma cells. In a few cases many of the cells, especially those about the central and sublobular veins, showed a hyaline type of degeneration, the cytoplasm staining deep red and either being homogeneous or containing hyaline droplets and granules.

A variable feature in half of the cases was the presence of cytoplasmic vacuolation, which appeared most marked just beneath the capsule of the liver, about the central vein (grade 1), or affecting the lobules diffusely in their entirety (grade 2). The vacuoles were usually largest around the central vein and diminished in size toward the periphery where they were minute and numerous and completely

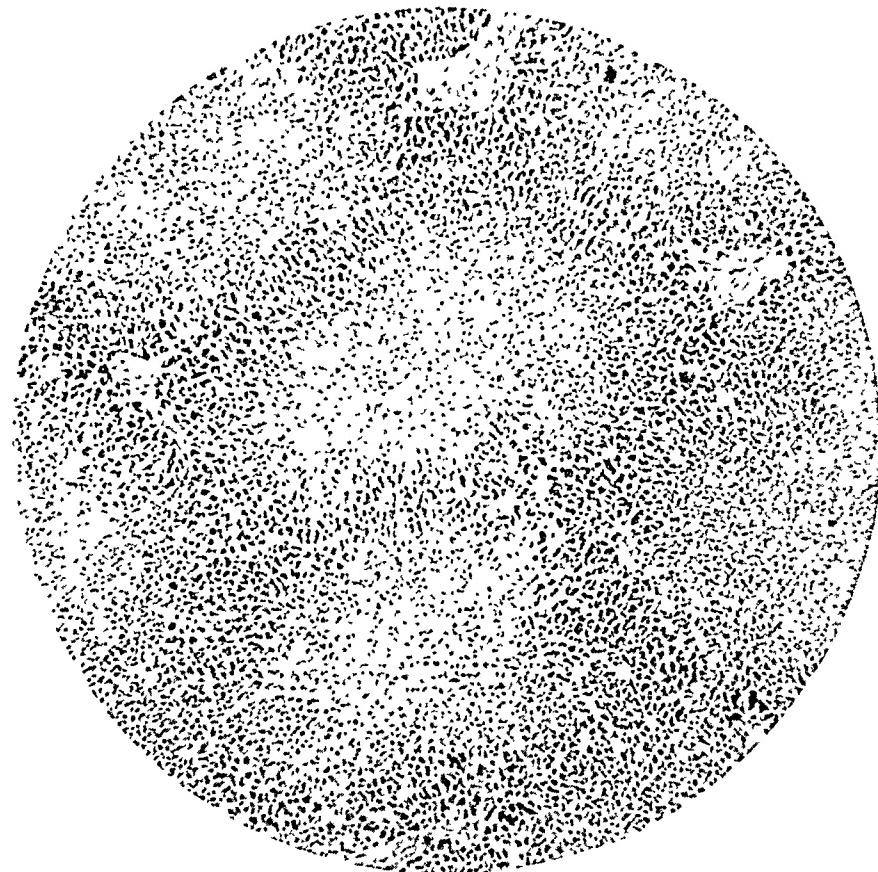
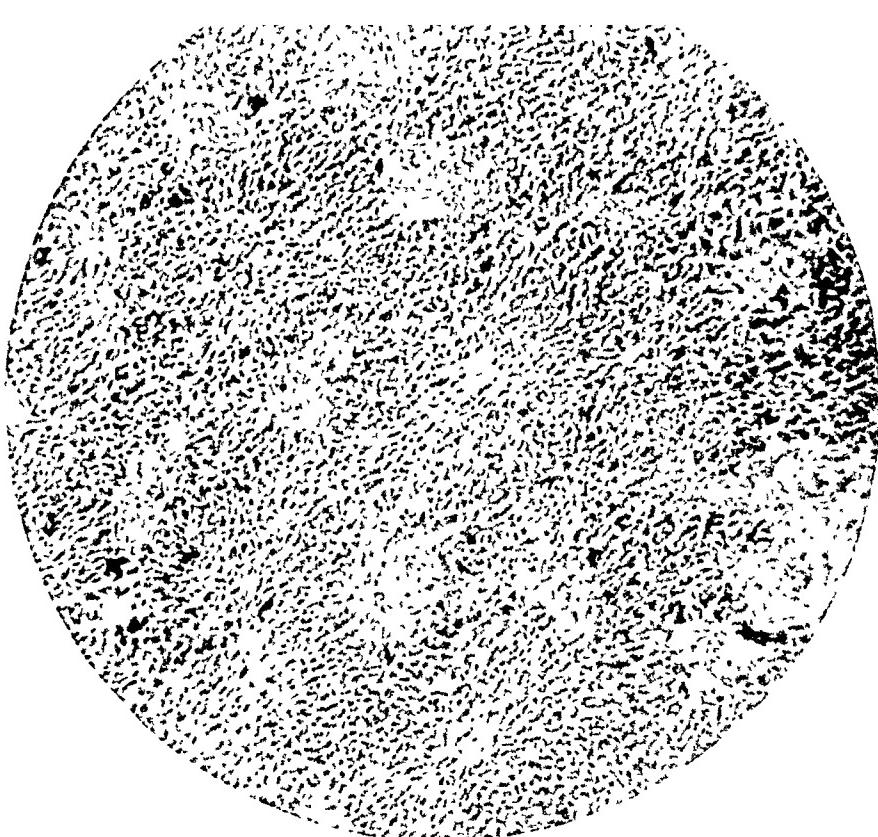


Fig. 2.—Extent of injury to the hepatic cells and disorganization of intralobular architecture;  $\times 40$ .

replaced the cytoplasm. Biliary pigment was sometimes contained within these vacuoles or concentrated about their circumferences. Hepatic cells isolated by prolongations of connective tissue appeared to have greater difficulty in disposing of their pigment and at times also showed cytoplasmic vacuolation. "Signet ring" forms were uncommon and, rarely, a large vacuole replaced two or three hepatic cells. None of the material was suitably fixed to react with glycogen stains, and only eight gross specimens were available from which to obtain sections to be stained for fat. Although it seems improbable that vacuoles of such size and morphology could be interpreted as due to anything but fat, this substance could not be demonstrated by any of the measures employed. It is possible that these vacuoles represented a type of hydropic degeneration. Nuclear vacuolation was present in six cases, occurring indiscriminately within the lobule, usually being inconspicuous and unrelated to cytoplasmic vacuolation. In the development of this phenomenon the chromatin material collected about the nucleolus, which migrated toward the nuclear membrane where it finally became concentrated, leaving the center more or less clear. The nucleus then swelled to two or three times its original size and appeared as a deeply basophilic nuclear ring, within which the nucleolus was sometimes discernible.

Focal midzonal areas of necrosis were present in fourteen cases and were essentially similar to those encountered under ordinary conditions of total biliary stasis. In a few instances the lesions were apparently enlarged as a result of decompression, and in two cases they eventuated in the formation of abscesses. The healing of these necroses appeared to be accomplished exclusively by regeneration and not by organization. Biliary necroses were present in three cases, involving single lobules in their outer thirds or in their entirety or, by extension, several adjacent lobules. When numerous and lying in close approximation to each other, they forced the intervening hepatic cords into more or less parallel rows composed of flattened cells. The pigment was either diffused or transported by phagocytic cells to the periphery of the lesion. No evidence of organization was observed about any of the areas of biliary necrosis. In three cases abscesses were present in relation to the biliary ducts or within biliary or focal midzonal areas of necrosis occupying at times several lobules and showing little evidence of liquefaction in their centers.

Monocytes occasionally seen in mitosis and swollen Kupffer cells containing engulfed pigment, erythrocytes, fragments of necrotic cells and other débris were observed in the sinusoids of many of the specimens. Focal areas of hyperemia were present in fifteen cases, and in these areas the sinusoids were markedly distended and closely packed with well preserved, hemolyzed or conglutinated erythrocytes. Hemorrhage was present in five of these, and the hepatic cells within the areas were atrophied and tended ultimately to disappear. Hyperemia was usually well marked in areas of necrosis, occasionally about the larger intrahepatic biliary ducts, and rarely generalized throughout the section. The accumulation of edema fluid in the perivascular tissue spaces appeared to be a prominent feature tending to compress the sinusoids and to render them bloodless. The sinusoidal reticular walls at times had become ruptured and fragmented in the inner third of the lobule, and rarely, red blood cells extruded into the perivascular tissue spaces. The arteries and arterioles regularly showed an increase in the medial musculature and, less frequently, subintimal proliferation of connective tissue. The branches of the portal vein were usually dilated and their walls thickened by perivascular fibrosis. Although thrombosis of blood vessels was present in several

preparations, it was not attended by infarction nor was it directly associated with necrotic areas in such a manner as to suggest an etiologic relationship.

The smaller biliary ducts which had proliferated while the liver was in a condition of total stasis collapsed following decompression, and their lining cells atrophied and finally vanished. The parietal sacculi tended to reappear in the walls of the larger biliary ducts, the lumens of which contained desquamated epithelial cells, leukocytes, erythrocytes and, in some instances, clumps of bacteria, probably representing an agonal invasion.

In amount and distribution the connective tissue in these livers was similar to that found under conditions of total stasis except that it was apt to be more edematous, was the seat of a greater infiltration of leukocytes and appeared compressed by the regenerating cells at the periphery of the lobule.

*Regeneration.*—Evidences of cellular regeneration were present in twelve cases and were characterized by nuclear fission and budding and by binucleation and multinucleation of hepatic cells. No mitotic figures were observed in any of the preparations studied. The nuclei in certain regenerating cells became elongated and constricted in the midportion. This indentation, slight at first, penetrated more deeply and resulted ultimately in the separation of the nucleus into two structures of equal size. From other nuclei a bud was formed which gradually enlarged to a size identical with that of the mother nucleus and then separated. Hypertrophic hepatic cells containing single large hyperchromatic nuclei were frequently noted. Regenerating cells were numerous in and about the necrotic areas surrounding the central vein and scattered sporadically either individually or in small groups throughout the lobule. Regenerative changes were seen about the areas of focal midzonal necrosis but not about the biliary necroses. Coincidentally with the formation of new hepatic cells the connective tissue became condensed at the periphery of the lobule. Groups of disorganized cells, in an apparent attempt to reestablish the continuity of the architectural pattern of the liver, put forth short cytoplasmic processes much like pseudopods which, however, often failed to unite with their parent cords. While regeneration of necrotic areas usually proceeded from without inward, it was also observed to occur simultaneously at both the inner and the outer borders of the necroses surrounding the central vein. Reconstruction of the necrotic areas was accomplished not only by regeneration but also by recovery of many of the constituent cells which, although badly damaged, were apparently still viable and capable of returning to normal. Conversely, however, many of the newly regenerated cells had undergone degeneration and necrosis.

*Comparative Studies.*—As a basis for this study it seemed most desirable to use cases of obstructive jaundice due only to carcinoma of the head of the pancreas. In these cases the condition of the liver and biliary ducts is usually normal prior to the onset of biliary stasis which is regularly complete and permanent until surgical relief is instituted. The influence of biliary stasis on regeneration is hard to evaluate in cases of obstruction of the common duct due to many other causes, in the presence of which it is extremely difficult to eliminate the possibility of previous or coexistent infection. However, the regressive changes in the hepatic cells and the disruption of the architectural pattern seem to be essentially similar in all the cases in which decompression, following biliary stasis, was effectual. These facts were determined by extending the study to include autopsy material from twenty-six additional cases of decompression of the biliary system, the cause of obstruction being carcinoma of the gallbladder (four cases), carcinoma of the duodenum (one case), carcinoma of the common hepatic duct (one case) and calculous cholecystitis (twenty-two cases). Large necrotic areas of obscure etiol-

ogy were present in two of the cases owing to calculous cholecystitis. Of additional particular interest was the presence, in six of these cases, of focal areas of sinusoidal thrombosis varying in size, shape, degree of parenchymal involvement and stage of development. They were usually situated immediately beneath the capsule of the liver and about the central and sublobular veins, being confined to a single or to several adjacent lobules. At first the lesion appeared as an irregularly shaped, poorly demarcated area of hemorrhage, within which could be noted a few isolated atrophied and necrotic hepatic cells. About the borders the sinusoids were hyperemic and the hepatic cells compressed. The red blood cells faded or conglutinated to form small, hyalin-like masses and a fine fibrin network was precipitated and later infiltrated by monocytes and polymorphonuclear leukocytes. Regeneration was well marked in and about these lesions.

#### COMMENT

If conditions are favorable following decompression the liver may return to a practically normal condition, with or without a variable amount of residual portal fibrosis. Evidence of regeneration may be marked in and about the areas of necrosis surrounding the central vein and in relation to the focal midzonal necroses, as well as sporadically within the lobule and at the periphery. MacMahon<sup>14</sup> observed extremely active regeneration in livers in which the continuity of hepatic cords and biliary capillaries was interrupted and in which biliary pigmentation was present about the somewhat swollen hepatic cells. No statement was made as to whether the cases dealt with were cases of obstruction of the common duct, of decompression or of some form of intrahepatic jaundice. The only reference found relative to hepatic regeneration under conditions of decompression is Bell's<sup>15</sup> work on cholecystostomy on dogs some time following previous ligation of the common bile duct. The rapidity of the regeneration depended roughly on the damage to be repaired and on the duration of the obstruction, usually being completed in from two to four months.

We were unable to find mitotic figures in the livers of any of our patients, but, in view of the opinions of others (Fishback<sup>16</sup> and MacMahon) with regard to hepatic regeneration, this may be of little significance, for figures indicative of mitotic division resulting in the formation of two or more nuclei in a single cell may have escaped observation. Rössle<sup>17</sup> was convinced that amitosis could play no rôle in the growth of tissue, and Doljanski<sup>18</sup> stated that he never saw a cell form in tissue cultures of the liver from which he could conclude

14. MacMahon, H. E.: Ztschr. f. mikr.-anat. Forsch. **32**:413, 1933.

15. Bell, L. P.: California & West. Med. **25**:503, 1926.

16. Fishback, F. C.: Arch. Path. **7**:955, 1929.

17. Rössle, R.: Handbuch der normalen und pathologischen Physiologie, Berlin, Julius Springer, 1927, vol. 14, p. 923.

18. Doljanski, L.: Arch. f. exper. Zellforsch. **11**:261, 1931.

that amitosis contributed to the multiplication of cells. However, recent investigators emphasize the importance of amitosis (Clara,<sup>19</sup> and MacMahon) with cytoplasmic division probably lagging behind (Schultz, Hall and Baker),<sup>20</sup> the number of binuclear cells being finally reduced to normal as a result of a prompt division of the hepatic cells in a series of crops (Ponfick).<sup>21</sup> An interesting question, as stated by Fishback, concerns the extent of participation of biliary ducts in the regeneration of hepatic cells. Marchand,<sup>22</sup> Ribbert<sup>23</sup> and Muir<sup>24</sup> stated that they found no proliferation of biliary ducts in the livers of human beings; they attributed the apparent proliferation to either degeneration or regeneration and rearrangement of the hepatic cells in an attempt to preserve their continuity. Meder,<sup>25</sup> Carraro,<sup>26</sup> Stroebe,<sup>27</sup> Barbacci,<sup>28</sup> MacCallum,<sup>29</sup> K. Hess,<sup>30</sup> O. Hess,<sup>31</sup> Miller and Rutherford<sup>32</sup> and Herxheimer and Gerlach,<sup>33</sup> on the contrary expressed a belief that biliary ducts do proliferate and, with the exception of Carraro, these authors thought that biliary ducts may serve as a source of new hepatic cells, at least under conditions of stress. They found the first evidence of regenerative activity to be present in the proliferating cells of the biliary ducts. Lieber and Stewart<sup>34</sup> found no regenerating hepatic cells in the livers of patients with complete and permanent stasis, despite the excessive and progressive proliferation of biliary ducts. In autoplasic hepatic transplants Cameron and Oakley<sup>35</sup> demonstrated that the hepatic cells and epithelium of the biliary ducts grow independently and show no tendency to unite with each other. As a result of the present study no evidence is adduced that following decompression the biliary ducts act as a source for the formation of new

19. Clara, M.: *Ztschr. f. mikr.-anat. Forsch.* **22**:145, 1930; **26**:45, 1931.

20. Schultz, E. W.; Hall, E. M., and Baker, H. V.: *J. M. Research* **44**:207, 1923.

21. Ponfick, E.: *Virchows Arch. f. path. Anat.* **230**:289, 1921.

22. Marchand, F.: *Beitr. z. path. Anat. u. z. allg. Path.* **17**:206, 1895.

23. Ribbert: *Arch. f. Entwicklungsmechn. d. Organ.* **18**:267, 1904.

24. Muir, R.: *J. Path. & Bact.* **12**:287, 1908.

25. Meder, E.: *Beitr. z. path. Anat. u. z. allg. Path.* **17**:143, 1895.

26. Carraro, A.: *Virchows Arch. f. path. Anat.* **195**:462, 1909.

27. Stroebe, H.: *Beitr. z. path. Anat. u. z. allg. Path.* **21**:379, 1897.

28. Barbacci, O.: *Beitr. z. path. Anat. u. z. allg. Path.* **30**:49, 1901.

29. MacCallum, W. G.: *Bull. Johns Hopkins Hosp.* **10**:375, 1902.

30. Hess, K.: *Virchows Arch. f. path. Anat.* **121**:154, 1890.

31. Hess, O.: *Beitr. z. path. Anat. u. z. allg. Path.* **56**:22, 1913.

32. Miller, J., and Rutherford, A.: *Quart. J. Med.* **17**:81, 1923.

33. Herxheimer, G., and Gerlach, W.: *Beitr. z. path. Anat. u. z. allg. Path.* **68**:93, 1921.

34. Lieber, M. M., and Stewart, H. L.: *Arch. Path.* **17**:362, 1934.

35. Cameron, G. R., and Oakley, C. L.: *J. Path. & Bact.* **38**:17, 1934.

hepatic cells. Indeed, under such circumstances, the regeneration of hepatic cells progresses rapidly, whereas the newly proliferated biliary ducts are undergoing involution. Furthermore, a large number of regenerating hepatic cells are present well within the lobule, distinctly separated from the biliary ducts by a zone of hepatic parenchyma at the periphery.

The material at our disposal was unsuited to the study of the hepatic changes dependent on the effects of a long continued anastomosis between the biliary tract and some portion of the gastro-intestinal tract. The experimental data pertaining to this condition in animals will be referred to later. In patients coming to autopsy shortly after decompression, the parenchyma of the liver was frequently extensively disorganized and degenerated, the regressive changes occasionally progressing to acute diffuse hepatic necrosis. It is not possible to compare these changes with those of acute yellow atrophy of the liver, since there is no unanimity of opinion as to the early picture of the latter lesion (Roman).<sup>36</sup> It would have been interesting to investigate the relationship between the effects of these destructive changes and the mechanism of jaundice, but, in this series of cases, functional studies following operation were unfortunately too meager to permit such a study. Available data indicate that, as a rule, the hyperbilirubinemia and clinical manifestations of jaundice steadily decrease in patients who recover following a release of biliary obstruction. Weir and Walters,<sup>13</sup> however, pointed out that a slow decline or a rise of bilirubin in the serum post-operatively indicates serious and progressive hepatic parenchymal injury. It has long been recognized (Judd and Lyons,<sup>37</sup> and Walters and Parham<sup>7</sup>) that if the draining bile becomes thin, pale and increased in volume the import is serious and indicates failing hepatic function. Walters, Greene and Frederickson<sup>38</sup> found that in patients with marked cholerrhagia following decompression, the concentration and total output of bilirubin were reduced and the characteristic constituents of the bile were practically entirely lacking, even though this deficient secretion was elaborated in considerable quantity. The regressive morphologic observations in certain of our cases permit the theory that the hepatic excretion of biliary pigment may be seriously impaired following operation, when the factor underlying the production of jaundice changes from an extrahepatic obstruction to an intrahepatic lesion, probably similar in all respects to that characteristic of acute diffuse necrosis (acute yellow atrophy of the liver).

36. Roman, B.: Arch. Path. **4**:399, 1927.

37. Judd, E. S., and Lyons, J. H.: Ann. Surg. **77**:281, 1923.

38. Walters, W.; Greene, C. H., and Frederickson, C. H.: Ann. Surg. **91**:686, 1930.

This prevailing and characteristic picture of extensive necrosis in combination with hepatic parenchymal dissociation apparently does not occur with any degree of frequency in conditions other than surgical decompression following total stasis. This conclusion is supported by observations made on a control series comprising twenty-four cases of total stasis due to carcinoma of the head of the pancreas without decompression and on thirteen instances of cholecystectomy or cholecystostomy in nonjaundiced patients with calculous cholecystitis. We also examined many livers obtained from patients dying as a result of a variety of conditions unrelated to the liver, gallbladder or biliary ducts, some of whom had been subjected to prolonged anesthesia. One of the control cases is of great significance. A patient with obstructive jaundice due to carcinoma of the common hepatic duct was subjected to cholecystogastrostomy which, of course, did not relieve the biliary stasis. He died eighteen hours following operation. The histologic preparations of the liver showed the typical picture of total stasis with complete absence, however, of the combined regressive changes in the cells and disruption of intralobular pattern so characteristic of the liver in cases of decompression as described. The assumption appears to be justified, therefore, that this picture is due not directly to prolonged anesthesia or operative procedures on the biliary tract, but to certain factors associated with decompression of an obstructed biliary system.

During total biliary stasis the hepatic blood pressure rises in an attempt to maintain adequate function of the hepatic cells in the face of the gradually increasing pressure which acts as an obstacle to the secretion or excretion of bile. The combination of acute dilatation of intrahepatic blood vessels occurring simultaneously with decompression of a hydrohepatotic liver results in sudden and marked alterations in pressure. The rapidity of induction of the changes may be one of the factors responsible for the production of marked parenchymal disorganization and may also account in part for the distention of sinusoids with corresponding atrophy of hepatic cell cords, edema of the perivascular tissue spaces, and fraying and tearing of sinusoidal reticular walls, particularly since the last mentioned lesions are most marked about the larger intrahepatic ducts where the effects of this force are probably greatest. Hemorrhage into the biliary conducting system may also be due to this sudden release in intraductal pressure, especially in patients with a hemorrhagic tendency already established. It is difficult to evaluate the importance of this factor in the production of widespread degeneration and other regressive changes, but pressure, as is well known, is a common cause of necrosis.

The chemical characteristics of the bile following the release of an obstruction of a common duct have been studied by a number of investi-

gators (von Czyhlarz, Fuchs and von Fürth,<sup>39</sup> Chabrol, Benard and Bariety,<sup>40</sup> Rosenthal, von Falkenhausen and Freund,<sup>41</sup> Greene, Walters and Frederickson,<sup>42</sup> Ravdin, Johnston, Riegel and Wright<sup>43</sup>). The formation of biliary acids, which may be partially or completely inhibited during total stasis, returns to normal relatively rapidly following decompression, if the liver has been not too seriously injured. Ravdin and his co-workers found a low calcium content and a high chloride level, which tended to fall in patients who recovered and to increase in those who died. The biliary salts were constantly absent in the liver bile when the common duct had been completely obstructed for a week or more, and they reappeared only after a variable period of from one to four weeks. The relationship which these chemical disturbances bear to the morphologic changes in the liver in cases such as we have described is at present unknown, although it seems logical to assume that the delayed reappearance of bile acids in the bile following decompression may be partly the direct result of the hepatic disorganization and regressive changes.

There is abundant experimental evidence to suggest that the creation of a biliary fistula or of an anastomosis between the gallbladder and any part of the gastro-intestinal tract is regularly attended by infection of the biliary passages and the liver (Gatewood and Poppens,<sup>44</sup> Lehman,<sup>45</sup> Horsley,<sup>46</sup> Beaver,<sup>47</sup> Gatewood and Lawton,<sup>48</sup> Gage<sup>49</sup> and Wangensteen<sup>50</sup>). The experience of Walters, Greene and Frederickson<sup>38</sup> with dogs with permanent biliary fistulas indicates that the development of cholangitis with associated cirrhotic changes in the liver may rapidly reduce the quantity of bile acids in the bile. The importance of infection as a serious complication in our cases of shorter duration is attested by the presence of fresh inflammatory changes in

39. von Czyhlarz, E.; Fuchs, A., and von Fürth, O.: Biochem. Ztschr. **49**: 120, 1913.

40. Chabrol, E.; Benard, H., and Bariety, H.: Bull. et mém. Soc. méd. d'hôp. de Paris **50**:992, 1926.

41. Rosenthal, F.; von Falkenhausen, M., and Freund, H.: Arch. f. exper. Path. u. Pharmakol. **111**:170, 1926.

42. Greene, C. H.; Walters, W., and Frederickson, C. H.: J. Clin. Investigation **9**:295, 1931.

43. Ravdin, I. S.; Johnston, C. G.; Riegel, C., and Wright, S. L.: J. Clin. Investigation **12**:659, 1933.

44. Gatewood, E. T., and Poppens, P. H.: Surg., Gynec. & Obst. **35**:445, 1922.

45. Lehman, E. P.: Arch. Surg. **9**:16, 1924.

46. Horsley, J. S.: South. M. J. **20**:669, 1927.

47. Beaver, M. G.: Arch. Surg. **18**:889, 1929.

48. Gatewood, E. T., and Lawton, S. E.: Surg., Gynec. & Obst. **50**:40, 1930.

49. Gage, I. M.: Proc. Soc. Exper. Biol. & Med. **28**:693, 1931.

50. Wangensteen, O. H.: Ann. Surg. **87**:54, 1928.

and about the larger ducts, in association with the biliary and focal midzonal areas of necrosis and elsewhere in the hepatic parenchyma in relation to the portal radicles and terminal biliary ducts. These lesions are probably the result of an ascending infection and may contribute to the production of vascular thrombosis and acute regressive changes in the liver.

#### SUMMARY

Following surgical decompression of an obstructed biliary system, hepatic pigmentation diminishes progressively in patients who survive the immediate effects of the operation, and the hepatic parenchyma tends to return to an approximately normal condition. This is accomplished by the recovery of many of the degenerated but still viable hepatic cells and also by regeneration. The latter is characterized by nuclear fission and budding and by binucleation and multinucleation of hepatic cells, without the appearance of mitotic figures. There is no evidence that hepatic cells arise from the biliary ducts, which rapidly involute following decompression. Lobular expansion subsequent to regeneration of hepatic cells results in compression and condensation of connective tissue at the periphery. The features of regeneration may be minimal or entirely absent in some livers in which the changes are those of a severe acute hepatosis occasionally complicated by hemorrhage and superimposed infection. Disruption of the intralobular architecture with disorganization and dissociation of hepatic cell cords occurs regularly. The necrosis in the inner third of the lobule often extends into the middle and outer thirds or involves the entire lobule, resulting occasionally in acute diffuse necrosis. Many of the focal midzonal and biliary necroses also enlarge and may form abscesses. In addition, the hepatic cells may be atrophied and distorted and at times show cytoplasmic and nuclear vacuolation. The recently regenerated hepatic cells may undergo degeneration and necrosis. The vascular changes include thrombosis, rupture of the sinusoidal reticular walls, hemorrhage, edema of the perivascular tissue spaces and hyperemia, either focal or general throughout the organ. Physical, chemical and infectious factors probably play an etiologic rôle in the pathogenesis of these lesions.

# CYTOTOLOGY OF PERITONEAL FLUID IN PARTIALLY HEPATECTOMIZED ANIMALS

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The cells of the peritoneal fluid have been the object of frequent and extensive studies ever since von Recklinghausen<sup>1</sup> studied the routes of absorption of materials injected into the peritoneal space. The literature is too extensive to review in this report, and the reader is referred to the rather complete review given by Webb.<sup>2</sup>

Earlier studies of these cells were concerned with their reaction to various substances injected into the peritoneum. Other studies were concerned with the genetic relationships of these cells to those of the blood stream, and still others with the origin of the cells of serous exudates in general. Cellular continuity and transformations in various inflammatory reactions have been extensively studied. The relation of these cells to immunity and their mobilization and transfer to other parts of the body in defense reactions have been followed.

More recently, in studies directed toward the protection of the peritoneum against infection, further interest has been centered on the cytologic character of the protective mechanism. Various vaccines administered prior to operation in the lower portion of the abdomen induce a protective effect which materially lowers the incidence of postoperative peritonitis.<sup>3</sup> Bargen<sup>4</sup> and his co-workers have found this procedure useful, and reported the results of the clinical applications of the method. The cytologic response to these vaccines is now being investigated, and Rixford<sup>5</sup> recently made a preliminary report on this work. Steinberg and Goldblatt<sup>6</sup> reported an experimental study on dogs in which peritoneal vaccination was carried out by the

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From the Division of Experimental Medicine, the Mayo Clinic.

1. von Recklinghausen, F.: *Virchows Arch. f. path. Anat.* **26**:172, 1863.

2. Webb, R. L.: *Am. J. Anat.* **49**:283, 1931.

3. Rankin, F. W., and Bargen, J. A.: *Arch. Surg.* **22**:98, 1931.

4. Bargen, J. A.: *Proc. Staff Meet., Mayo Clin.* **8**:581, 1933.

5. Rixford, E. L.: *Proc. Staff Meet., Mayo Clin.* **8**:586, 1933.

6. Steinberg, Bernhard, and Goldblatt, Harry: *Surg., Gynec. & Obst.* **57**:15, 1933.

injection of a suspension of dead organisms in a solution of tragacanth. A marked increase in the total cell count to 153,000 per cubic millimeter of peritoneal fluid was observed within ten hours. Differential cell counts were not recorded, although the authors regarded the polymorphonuclear neutrophil as the predominant cell for the first forty-eight hours, after which there was an appreciable increase in the number of the mononuclear cells. Control data on both the total and the differential peritoneal cell count prior to the injection of the vaccine would facilitate the interpretation of these results.

This report is concerned with changes induced in the total and differential peritoneal cell count in animals from which a considerable portion of the liver had been removed. Recently, one of us (Montgomery<sup>7</sup>), in a study of the peritoneal cells of various laboratory animals, demonstrated that the total number of cells per cubic millimeter of fluid in the white rat was normally in excess of 100,000. This was an advance over the previous work on this subject, which had concerned itself with the determination of percentages only and suggested the work which is now being reported. Using the technic described previously, it was possible to determine, within the limits of error, the number of cells per cubic millimeter in the peritoneal fluid of animals. Since these cells increase enormously in the presence of various irritants and, furthermore, since they migrate to other regions of the body to combat infections, we were interested to know the effect of surgical intervention in the upper portion of the abdomen on the number of cells per unit of fluid in the peritoneal space. Partial removal of the liver, which is readily accomplished in rats, was selected as the operative procedure. The liver, of course, was not considered a source for these exudative cells, but we believed that the operation incident to its removal might not be without effect. The white rat was selected for the study.

#### PROCEDURE

Ten healthy male white rats, 6 months of age and weighing between 200 and 225 Gm., were selected for this study. They were in good physical condition, so far as could be determined. To secure adequate control data, five samples of peritoneal fluid were taken from each animal during three weeks prior to the operation. Both total and differential cell counts were made on these fluids. The smears were stained with Wright's stain, and 200 cells were counted from each smear. From five of the animals about 75 per cent of the liver was removed aseptically by a technic described in a previous article.<sup>8</sup> In three animals none of the liver was removed, but a small portion of one lobe was excised and transplanted into the peritoneal space. This served as a control for the hepatectomized

7. Montgomery, L. G.: Proc. Staff Meet., Mayo Clin. 7:589, 1932.

8. Higgins, G. M., and Anderson, R. M.: Arch. Path. 12:186, 1931.

group, in which a small remnant of hepatic tissue always remained adjacent to the level of the ligature. In two of the rats simple laparotomy alone was performed. Samples of blood and peritoneal fluid were taken on the third day after operation and at frequent intervals thereafter for ten weeks.

### RESULTS

Since the morphology of the cells of the peritoneal fluid of the white rat has been adequately described, we shall restrict our consideration to an analysis of the total and relative number of cells present before and after partial hepatectomy.

On the basis of the fifty control counts that were made on the ten animals prior to operation, a mean total cell count of  $127,360 \pm 9,150$  per cubic millimeter of peritoneal fluid was determined. There was

TABLE 1.—*Total Number of Peritoneal Cells per Cubic Millimeter Before Operation (Control Counts)*

Animal	Total Number of Cells (Average of Five Counts)*
1.....	$103,400 \pm 17,100$
2.....	$82,900 \pm 4,600$
3.....	$147,300 \pm 9,300$
4.....	$84,400 \pm 8,000$
5.....	$99,300 \pm 6,200$
6.....	$205,300 \pm 6,500$
7.....	$190,300 \pm 11,500$
8.....	$149,800 \pm 6,600$
9.....	$116,400 \pm 7,900$
10.....	$94,500 \pm 7,100$

\* The average for the entire series (50 counts) was  $127,360 \pm 9,150$ .

a wide variation in the mean total cell count of the peritoneal fluid of the individual animals (table 1). The lowest mean count recorded was  $82,900 \pm 4,600$ , and the highest,  $205,300 \pm 6,500$ . It will be noted from this table that the probable errors, except in the case of animal 1, were not large, and thus there was considerable regularity in the counts determined. In rats with a high peritoneal cell count the count remained consistently high, and in those with a lower count it remained consistently low, during the period in which control counts were taken. These differences are significant statistically, but we offer as yet no explanation why counts for one animal should be double those found for another. We believe that the variation may be correlated, however, with the presence of absorptive phenomena in the peritoneum.

The differential peritoneal cell counts, on the other hand, revealed more uniformity for the whole group. We did not attempt to distinguish the clasmacytotes from the monocytes and large lymphocytes, but placed them in one group designated as mononuclear cells. The

small lymphocytes were classified as lymphocytes. The eosinophilic leukocytes were easily identified, as were the large mast cells. We noted only a small percentage of polymorphonuclear neutrophils in our series, which indicated that a pathologic condition may have existed, for it is generally conceded that these cells do not occur in serous fluids unless infection is present. This condition was transient, however, for soon after recovery from the operation neutrophils were not observed in the smears.

The average differential count of the 10,000 cells which served as a control is shown in table 2. Our figures agree essentially with those

TABLE 2.—*Percentage of Peritoneal Cells in All Ten Animals Prior to Operation, (Fifty Counts, 10,000 Cells)*

	Per Cent
Mononuclears.....	$56.90 \pm 0.67$
Lymphocytes.....	$3.94 \pm 0.24$
Eosinophils.....	$32.12 \pm 0.78$
Neutrophils.....	$4.20 \pm 0.67$
Mast cells.....	$2.82 \pm 0.16$

TABLE 3.—*Total Number of Peritoneal Cells per Cubic Millimeter After Partial Removal of the Liver*

Time After Operation	Average Count (Five Animals)
3 days.....	$10,250 \pm 1,190$
5 days.....	$10,370 \pm 750$
14 days.....	$17,600 \pm 3,100$
28 days.....	$39,600 \pm 4,600$
42 days.....	$61,000 \pm 8,500$
56 days.....	$69,800 \pm 7,800$
4 months.....	$29,800 \pm 2,000$

of Webb, except that he did not encounter neutrophils in his smears and found a much larger mast cell count ( $7.36 \pm 0.38$  per cent). The lowest mean percentage of mononuclear cells recorded for any animal was  $52.84 \pm 1.03$ , and the highest,  $62.30 \pm 1.70$ . The difference, together with its probable error, is  $9.46 \pm 1.98$ , indicating that there is a slight significant difference between these two percentages.

In the animal in which the mononuclear count was low the lymphocyte count was also low, but the eosinophilic leukocyte count was high. Accordingly, in these groups the difference between the high and low differential counts was about four times the probable error of the difference, thus bordering on the significant. We believe, however, that the figures in table 2 represent fairly well the control data prior to operation.

The data assembled from the total peritoneal cell counts after partial hepatectomy have been condensed into table 3, and the data obtained from the control group in which simple laparotomy or laparotomy with implantation of liver was performed have been condensed into table 4. It is observed that, on the third postoperative day, the mean count recorded for the five animals from which the liver had been removed had fallen to  $10,250 \pm 1,190$  (table 3), whereas the count for the five control animals was  $129,300 \pm 20,000$  (table 4). There was no essential difference between the preoperative and postoperative counts in control animals from which the liver had not been removed, whereas a marked fall from a mean of 127,360 to one of 10,250 occurred in the animals subjected to partial removal of the liver. It was evident, too, that there was no effect on the number of peritoneal cells in the fluid of animals that had had autogenous implants of liver into the peritoneum.

TABLE 4.—*Total Number of Peritoneal Cells per Cubic Millimeter After Laparotomy and Implanting Small Portion of Liver into Peritoneum*

Time After Operation	Average Count (Five Animals)
3 days.....	$129,300 \pm 20,000$
5 days.....	$134,000 \pm 16,000$
14 days.....	$173,100 \pm 9,500$
28 days.....	$123,400 \pm 13,000$
42 days.....	$151,800 \pm 20,500$
56 days.....	$160,100 \pm 13,500$
4 months.....	$186,300 \pm 6,200$

The postoperative total cell count appeared to bear no relation to the level of the preoperative count; the mean control count of  $147,300 \pm 9,300$  (table 1) in animal 3 dropped on the third day after operation to 4,500 per cubic millimeter, whereas the lower initial control cell count of  $84,400 \pm 8,000$  in animal 4 dropped to only 14,000. The mean peritoneal cell count, taken at intervals during recovery and restoration of the liver, remained low and never fully regained the level established as the preoperative control count. The mean total count taken four months after operation— $29,800 \pm 2,000$ —was exceedingly low and indicated that great variation from the normal peritoneal cell count might be expected in animals in which the restored liver had assumed special relations somewhat different from those of the normal organ. The data of postoperative cell counts for the control series (table 4) revealed no marked deviation from the preoperative control levels. The counts, as a rule, were higher than the mean preoperative control count, but there were no marked changes such as were found in the animals from which the liver had been removed.

The differential peritoneal counts during the postoperative period did not vary greatly from the control (fig. 1), except the polymor-

phonuclear neutrophil counts, which, during the first three days after operation, rose to a percentage of  $25.00 \pm 3.81$ . The percentage of mast cells remained the same, whereas the percentage of mononuclear leukocytes and eosinophils dropped to  $35.40 \pm 4.56$  and  $26.80 \pm 4.12$ ,

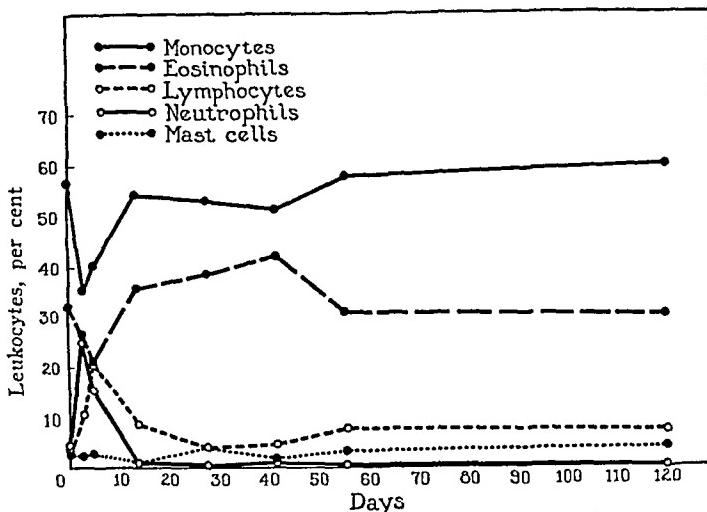


Fig. 1.—Differential peritoneal cell count after partial removal of the liver.

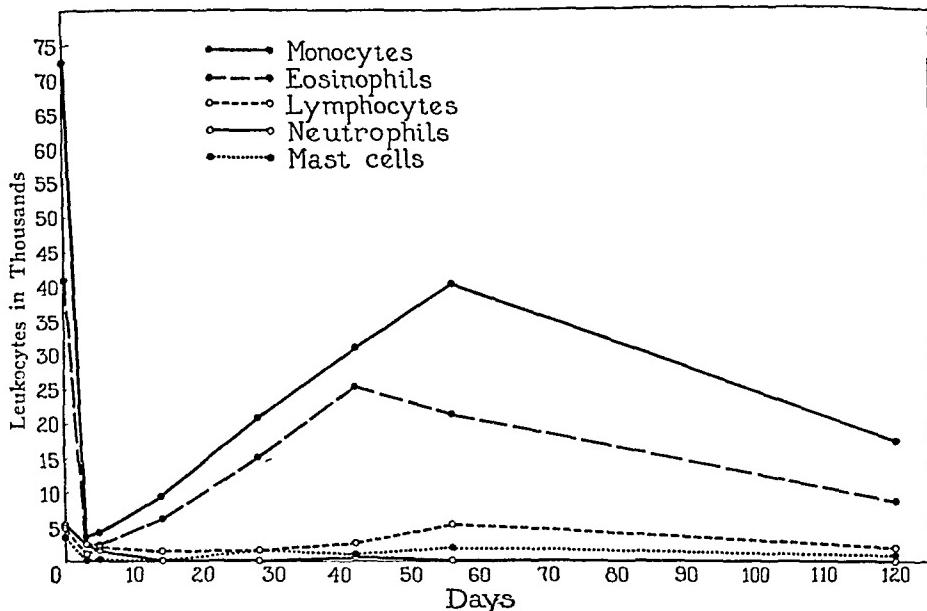


Fig. 2.—Total number of different kinds of peritoneal cells after partial removal of the liver.

respectively. Computing the number of cells present on the basis of the differential count (fig. 2), it appears that the mononuclear cells had dropped from 72,000 to 3,000, the eosinophils from 40,000 to 2,000 and the mast cells from 3,500 to 2,300. Although the percentage of

neutrophils had increased greatly, the actual number of neutrophils had fallen from 5,300 to 2,600 during the three day period. On the fourteenth day after operation a more nearly normal relative distribution of cells was restored, although the mean total cell count was low — $17,600 \pm 3,100$ . Although there was a gradual increase in the total number of cells per cubic millimeter of peritoneal fluid up to the eighth week, the relative distribution remained essentially unchanged. This would seem to indicate that whatever factors determined the cell content of the peritoneal fluid affected all types of cells equally.

There were no significant changes in the relative distribution of the cells in the fluid of the five control animals following operation. There was a transient rise in eosinophilic leukocytes and a correspond-

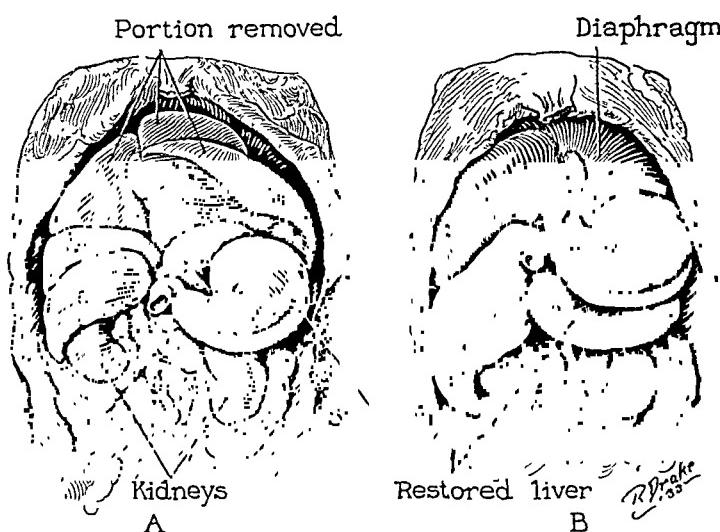


Fig. 3.—A, normal liver in peritoneum of rat; B, restored liver in peritoneum of rat after partial hepatectomy.

ing fall in the percentage of mononuclear cells, but by the fourteenth day a normal distribution was regained.

In the normal rat the convex surface of the liver is closely applied to the serosal surface of the diaphragm (fig. 3A). As a result of partial hepatectomy, whereby entire hepatic lobes are removed, a considerable portion of the surface of the diaphragm is exposed directly to the peritoneal cavity and its contents. As restoration of the liver occurs and the three small remaining hepatic lobes greatly increase in size, the surface of the diaphragm is again partially covered by the encroaching liver. However, even when restoration is complete and a liver equal to or greater than the normal has been restored, a considerable segment of the diaphragm remains in more or less direct continuity with the peritoneal space (fig. 3B). These changes in the spatial relations of

the diaphragm and liver following partial hepatectomy and the consequent restoration of the liver may explain the low peritoneal cell counts which we obtained. The diaphragm is the organ through which absorption from the peritoneal space most readily occurs. We are certain that these low cell counts are not due to factors of dilution.

#### SUMMARY

A study of the cells of the peritoneal fluid of the albino rat was made to determine the effect of partial hepatectomy on the total number of these cells and their differential relationships. It was found that a marked decrease in the total number of the cells occurred, whereas no significant change was found in the relative numbers of the different types. In a control group of animals in which simple laparotomy was performed, either with or without implantation of hepatic tissue into the peritoneal cavity, significant changes were not found.

# PHAGOCYTIC BEHAVIOR OF INTERSTITIAL CELLS OF BRAIN PARENCHYMA OF ADULT RABBIT TOWARD COLLOIDAL SOLUTIONS AND BACTERIA

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With the separation by Rio-Hortega<sup>1</sup> of the third element of Cajal into two groups, the microglia and the oligodendroglia cells, believed by him to possess distinct functional, morphologic and staining properties and a separate origin, the modern knowledge of these cells may be said to have begun. Hortega concluded that not only do the microglia cells possess the two properties essential for the proper functioning of a macrophage, viz., mobility and phagocytosis, but collectively they represent broadly the so-called reticulo-endothelial system locally in the brain. In spite of his investigations and those of his pupils, there still exists a controversy as to whether the microglia cells are the sole source of cerebral macrophages or whether the latter have a double origin from microglia cells and astrocytes. Only a few experiments have been conducted to demonstrate the phagocytosis of vital dyes by microglia cells, and none have been made on the behavior of these cells toward bacteria. The discrepancies in the results obtained by different workers are due primarily to inadequate technic or to differences in interpretation. Since the storage of true colloidal solutions and bacteria is unquestionably bound up with the process of phagocytosis, these can be employed to determine the phagocytic properties of the various interstitial cells of the rabbit's brain.

## REVIEW OF THE LITERATURE

Testa,<sup>2</sup> Cavallaro<sup>3</sup> and Gozzano<sup>4</sup> demonstrated ingested vital dyes in microglia cells. By the use of gum arabic to weaken the intensity of silver impregnation, Bolsi<sup>5</sup> noted granules of trypan blue in microglia cells. Russell<sup>6</sup> observed the affinity of transitional microglia cells for trypan blue in incompletely impregnated and toned sections of aseptically

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1. del Rio-Hortega, P.: Arch. de neurobiol. **11**:212, 1921.
2. Testa, M.: Folia med. **14**:725, 1928.
3. Cavallaro, V.: Pathologica **19**:11, 1927.
4. Gozzano, M.: Riv. di neurol. **1**:377, 1928.
5. Bolsi, D.: Riv. di pat. nerv. **37**:1, 1931.
6. Russell, D.: Am. J. Path. **5**:451, 1929.

injured cerebral tissue of rabbits. I found that the exact repetition of Russell's technic yielded, in general, unsatisfactory results, for not only were the cytoplasmic microglial processes very coarsely granular but they could not always be distinguished with certainty from the similarly coarse granular elements of the ground substance. Beletzky and Garwaki<sup>7</sup> observed the adsorption of ferric saccharate by mesoglia cells, but they did not state whether the ferric saccharate was in a true colloidal state; this is unfortunate, for it is well known that there are samples of saccharated or nonsaccharated oxide of iron which yield clear brown solutions free from suspended particles. Bratianu and Llombart<sup>8</sup> stated that the microglia cells do not possess the power of "fixing" colored colloidal solutions either in the normal or in the pathologic state and that macrophages arise not only from microglia cells but from oligodendrocytes and adventitial cells as well.

In spite of their close resemblance to macrophages in tissue cultures, Costero<sup>9</sup> claimed to have cultivated microglia cells in vitro from the brains of from 2 to 3 month human embryos, of from 7 to 20 day chick embryos and of new-born guinea-pigs and to have demonstrated the capacity of these cells for ingesting lithium carmine. Wells and Carmichael<sup>10</sup> observed in sections of cultures of embryo chick brains specifically impregnated for microglia cells wandering cells interpreted by them as being similar to microglia cells and capable of taking up in vitro neutral red and trypan blue. Mihálik<sup>11</sup> likewise noted in chick embryo brain cultures migrating and wandering cells which he identified as macrophages and as the third element of Cajal. It is evident, therefore, that until microglia cells of unquestioned identity are grown in vitro, preferably, if possible, in tissue culture consisting only of these cells, it is idle to speak of the phagocytic power of microglia cells for vital dyes in vitro.

On the basis of purely morphologic observations, the view has long been advanced that neuroglia cells are capable of producing macrophages (Merzbacher,<sup>12</sup> Alzheimer,<sup>13</sup> Creutzfeldt and Metz<sup>14</sup>). Alz-

7. Beletzky, W., and Garwaki, N.: Ztschr. f. d. ges. Neurol. u. Psychiat. **132**:474, 1931.

8. Bratianu, S., and Llombart, A.: Ann. d'anat. path. **62**:849, 1929.

9. Costero, I.: Ztschr. f. d. ges. Neurol. u. Psychiat. **132**:371, 1931.

10. Wells, A. Q., and Carmichael, E. A.: Brain **53**:1, 1930.

11. Mihálik, P.: Anat. Rec. **54**:157, 1932.

12. Merzbacher, L., in Nissl, F., and Alzheimer, A.: Histologische und histopathologische Arbeiten über die Grosshirnrinde, Jena, Gustav Fischer, 1909, vol. 3, p. 1.

13. Alzheimer, A., in Nissl, F., and Alzheimer, A.: Histologische und histopathologische Arbeiten über die Grosshirnrinde, Jena, Gustav Fischer, 1921, vol. 3.

14. Creutzfeldt, H. G., and Metz, H.: Ztschr. f. d. ges. Neurol. u. Psychiat. **106**:18, 1926.

heimer termed the macrophages derived from any type of astrocyte "ameboid glial cells." Later, Rosenthal<sup>15</sup> interpreted these cells as a type which suffered regressive changes and denied to them any phagocytic properties. Rio-Hortega and Penfield<sup>16</sup> observed no suggestion of the transformation of astrocytes into macrophages at the borders of experimental cerebral traumatic lesions in rabbits.

Nevertheless, this classic theory still attracts many adherents. Creutzfeldt and Metz<sup>14</sup> concluded after a study of fixed sections of diseased human cerebral tissue that the astrocytes contribute in part to the formation of macrophages. However, they differed from Alzheimer, who stated that the phagocytic function was common to all types of glial cells in maintaining that mobile macrophages originate from microglia cells and fixed macrophages from astrocytes. This concept of a fixed scavenger cell is not entirely acceptable in that it denies to the cells one of the two fundamental properties requisite for their function, the power of locomotion.

Even the application to this problem of vital staining and tissue cultures has led to contradictory results. Thus Goldmann<sup>17</sup> in his pioneer study of vital staining of the central nervous system observed more or less diffuse coloring of the astrocytes after cerebral subarachnoid injection of trypan blue in rabbits. Aside from the obvious fact that he was dealing with injured or dead cells, his observations must be discounted because of the complicating toxicity of the dye, as indicated by the fact that the initial injection was followed by an immediate reaction consisting of muscular spasms, convulsions and coma and death occurred nine hours later. Rachmanow<sup>18</sup> and Macklin and Macklin<sup>19</sup> observed storage of trypan blue in astrocytes about cerebral injuries in rabbits and rats, respectively. Mandlestanim<sup>20</sup> concluded that astrocytes "activated" by trauma are well stained by trypan blue. Recently, Roussy, Lhermitte and Oberling<sup>21</sup> claimed to have demonstrated trypan blue granules in astrocytes in rabbit brains directly exposed to radium. They evidently ignored one of the accepted signs of life, death or injury of the cell, for clearly the astrocytes represented in figure 12 of their article are either dead or injured, as shown by the diffuse or granular trypan blue

15. Rosenthal, S., in Nissl, F., and Alzheimer, A.: *Histologische und histopathologische Arbeiten über die Grosshirnrinde*, Jena, Gustav Fischer, 1913, vol. 6, p. 69.

16. del Rio-Hortega, P., and Penfield, W.: *Bull. Johns Hopkins Hosp.* **41**:278, 1927.

17. Goldmann, E. E.: *Abhandl. d. k. Preus. Akad. d. Wissensch.*, 1913.

18. Rachmanow, A.: *Folia neuro-biol.* **7**:750, 1913.

19. Macklin, C. C., and Macklin, M. T.: *Arch. Neurol. & Psychiat.* **3**:353, 1920.

20. Mandlestamm, A.: *Ztschr. f. d. ges. exper. Med.* **62**:471, 1928.

21. Roussy, G.; Lhermitte, J., and Oberling, C.: *Rev. neurol.* **1**:878, 1930.

staining of both the cytoplasm and the nuclei. Hence, they were not dealing with a true phagocytic process, and their observation is not valid. Furthermore, Russell,<sup>6</sup> Bolsi<sup>5</sup> and Bratianu and Guerriero<sup>22</sup> were unable to confirm their observations in respect to astrocytes in either traumatic or experimentally induced lesions in the rabbit's brain. Verne<sup>23</sup> observed in the second phase in the life of cultures of nerve tissue from 6 to 10 day chick and rat embryos emigrating cells interpreted as neuroglia cells which had the power of ingesting lithium carmine in vitro and of transforming themselves into macrophages. Examination of his drawings compels the conclusion that these "neuroglia cells" closely resemble true macrophages and are probably true wandering cells.

Prujjs,<sup>24</sup> Ferraro and Davidoff<sup>25</sup> and Cramer and Alpers<sup>26</sup> concluded that the oligodendroglia cells contribute to the formation of macrophages, although this is denied by Rio-Hortega, Penfield and others. According to Cramer and Alpers<sup>26</sup> in experimental secondary degeneration of the spinal cord of rabbits, the oligodendroglia cells are the first cells to function as myeloclasts, and they later serve as both myeloclasts and myeloblasts in conjunction with the microglia cells.

#### EXPERIMENTAL TECHNIC

The rabbit was selected as the experimental animal, principally because of the ease of silver impregnation of the microglia and other interstitial cells of its nervous parenchyma. The animals were laboratory-bred males, free from disease and weighing from 4½ to 5 pounds (2 to 2.25 Kg.).

They were anesthetized by ether, and a hole was bored with a hand drill through the right parietal bone 3 cm. to the right of the superior longitudinal venous sinus in order to avoid hemorrhage, visual disturbances and motor paralysis.

A puncture from 1 to 1.2 cm. in depth was made in the right parietal lobe with a sterile cold lumbar puncture needle in which the stylet was inserted. The injury served two useful purposes: (1) facilitation of the passage of colored semicolloidal and colloidal solutions into the nervous parenchyma by breaking the continuity of the hemato-encephalic barrier and by opening the smaller blood vessels and capillaries, thus establishing direct, physical contact of the surface of the interstitial cells with the dyes; (2) stimulation of an interstitial cellular reaction by the damaged brain tissue allowed to remain in situ. The operation was well supported. Not a single rabbit showed signs of infection either of the meninges or of the brain proper, and in not a single instance did paralysis follow.

*A. Trypan Blue.*—The dye in a concentration of 2 per cent was suspended in sterile triple-distilled water. A maximum concentration of 2 per cent was used in order to stain the wound tract and the neighboring brain tissue more deeply, if possible, than is the case with the 1 per cent suspension that has usually been employed by others. To demonstrate this, cerebral wound tracts were produced in a trial

22. Bratianu, S., and Guerriero, C.: Arch. d'anat. micr. **26**:337, 1930.

23. Verne, J.: Compt. rend. Assoc. d. anat., 1930.

24. Pruijs, W. M.: Ztschr. f. d. ges. Neurol. u. Psychiat. **108**:298, 1927.

25. Ferraro, A., and Davidoff, L. M.: Arch. Path. **6**:1030, 1928.

26. Cramer, F., and Alpers, B. J.: Arch. Path. **13**:23, 1932.

series of four rabbits which were then given 10 cc. of the suspension intravenously daily for seven days, while four rabbits with similar wounds received the same amount of dye over the same period but in a 1 per cent concentration. Sections from the brains and livers of both sets of animals counterstained with aluminum carmine and examined under a Zeiss comparison microscope revealed qualitatively a definitely larger number of stored dye particles in the cerebral macrophages and the reticulo-endothelial cells of the liver in the rabbits given the more concentrated suspension. At the same time, the toxicity of the dye used was observed; except for an almost negligible loss of weight, these heavy doses were not followed by toxic symptoms or signs.

Immediately before use, the solution was filtered to remove any aggregates capable of causing death by embolism and warmed to 37 C.

The animals in which trypan blue was used were separated into three groups according to the route of injection: (1) intravenous, (2) intra-arterial and (3) intraventricular.

**Intravenous Injection:** Ten rabbits received 10 cc. of a 2 per cent solution of the dye daily immediately after operation and five or six hours before death. Those surviving for two months received a total quantity of 600 cc., an amount certainly sufficient to produce at least a partial blockade of the so-called reticulo-endothelial system. Such a blockade would permit more dye than usual to reach the nerve cells capable of storing it. The animals were killed with chloroform after twelve, twenty-four, thirty-six, forty-eight and seventy-two hours and five, ten, twenty-four, forty and sixty days, respectively. The entire brain was removed immediately after death.

The puncture stab in the brain removed after twenty-four hours appeared as a slightly depressed, dark blue necrotic area, 2 mm. in diameter. The wound was frequently adherent to the dura and hemorrhagic at the margins and showed a moderate blue color in the necrotic center surrounded by a wider light sky-blue zone from which the color faded imperceptibly into the normal tissue. The dye was fixed principally at the site of the injured and dead tissue, and in contrast with the macroscopic appearance observed in the brains into which india ink and colloidal iron hydroxide were injected, it was highly diffused. Examination of the surface of the twenty-four hour brain with a Zeiss 20 X compensating ocular lens showed a zone of visible coloration extending 8 mm. from the central wound tract. Depending on the period of survival, the brain showed a progressively increasing intensity of blue staining in the central necrotic and hemorrhagic area, and the zone of reaction showed an increased width extending up to 13 mm. from the outer margin of the tract.

**Intra-Arterial Injection:** In order to bring a maximum quantity of dye to the injured brain before its deposit in other organs, intra-arterial injections were made in four rabbits. Immediately after traumatic injury of the parietal area of the brain, the carotid artery in the neck was exposed, and 10 cc. of a 2 per cent solution of trypan blue was introduced slowly into one of the smaller branches of the artery, which was then tied off. This procedure was repeated every other day, different branches of the same carotid artery being used. A rabbit was killed with chloroform every other day up to eight days after operation. The fourth rabbit, allowed to survive eight days, received therefore a total quantity of 40 cc.

Although unquestionably superior to injections into an auricular vein, injections into the carotid artery could not be carried out more often chiefly because of hemorrhage. Examination of the brains of the animals that had received intra-arterial injections showed a decidedly deeper intensity of staining than was present in

the brains of the rabbits given intravenous injections, but the distribution of the dye was essentially the same in both sets of animals.

**Intraventricular Injection:** Six rabbits without previous cerebral injury were given injections into the ventricles outside of the hemato-encephalic barrier so that an abundant amount of dye could be made accessible to the interstitial cells. After exposure of the dura, a syringe to which was attached a 2 mm. gage needle was gently passed through the gray and the white matter, and as soon as the needle appeared to be free in the ventricle, 2 cc. of a 2 per cent solution was slowly introduced. This was repeated every third day. The animals survived without any ill effects and were killed with chloroform from three to eighteen days after the initial injection. The brain tissue damaged by the passage of the needle was carefully removed because the dye penetrating from the ventricle into the gray and white matter had been taken up by degenerated or dead nerve cells, with the consequent production of artificial phagocytosis. Inspection of the brains revealed deep blue staining of the subependymal nerve tissue for from 1 to 1.5 cm. from the ependymal lining, with gradual but only moderate fading of color near the leptomeninges.

The rabbits given intravenous injections presented at autopsy an intense, deep blue staining of the skin, eyes, ears, liver, spleen and kidneys. The dura mater and the choroid plexus were deep blue.

**B. India Ink.**—Higgins' waterproof black drawing ink containing particulate carbon in colloidal suspension was selected as the source of the carbon particles for the phagocytic test. The ink was diluted with an equal volume of sterile triple-distilled water and immediately before use was warmed to body temperature.

Traumatic injury of the parietal lobe was produced as previously described in ten rabbits, which were then given intravenous injections of 10 cc. of the diluted ink daily for the first four days and then every other day until the end of eight weeks. The animals were killed after the same intervals and by the same technic as the rabbits into which trypan blue was injected. The stab wounds were from dark brown to almost black and surrounded by a zone of reaction from 2 to 4 mm. in width which was from light to moderate brown with sharp fading into the normal nerve tissue. So far as gross inspection permitted, it was evident that the ink possessed extremely slight, if any, power of diffusion.

**C. Colloidal Ferric Hydroxide.**—A colloidal suspension of ferric hydroxide was prepared as follows: Ferric chloride was purified by adding 95 per cent alcohol, drop by drop, until a precipitate was formed, and the solution was brought to a boil. The precipitate was filtered and dried. The presence of chlorine was tested for by the addition of 2 cc. of silver nitrate to the original filtered solution; a cloudy appearance indicated the presence of the chlorine ion.

To a freshly prepared half-saturated solution of purified ferric chloride (chemically pure) a 2 per cent normal solution of ammonium carbonate (chemically pure) was added in drops and with stirring until the precipitate formed just ceased to be dissolved. The solution was filtered if necessary and then dialyzed in a parchment tube against distilled water until only a trace of chlorine was detected. Gelatin was used as a protective colloid in an amount equal to about 0.073 per cent by weight. The solution was sterilized at 15 pounds' (6.75 Kg.) pressure for fifteen minutes in a steam autoclave. After sterilization, it was determined that the suspension contained 0.01147 Gm. of iron per cubic centimeter of liquid. The final solution was reddish brown.

A ferric hydroxide solution was also prepared as follows: Twenty cubic centimeters of a 2 per cent solution of ferric chloride (chemically pure) was gradually

added to 200 cc. of boiling water, and reddish-brown ferric hydroxide was produced by hydrolysis.

By repeated trial it was determined that the final preparation by either method was chemically inert and completely nontoxic for rabbits and that it was a complete suspension of submicronic particles. The serum of a rabbit in a dilution of 1:20 did not destroy its colloidal state in vitro, and therefore it was not surprising that it possessed the virtue of not producing minute emboli in the lung, as is so often the case with colloidal iron.

Before use, the liquid was allowed to stand for one hour to permit the suspended particles of iron to settle, and the supernatant clear fluid was carefully decanted. The concentrated suspension, each cubic centimeter of which contained 0.4588 Gm. of iron, was warmed to body temperature. Ten rabbits were given 20 cc. intravenously daily for two months after cerebral injury. They were then killed after twelve, twenty-four, thirty-six, forty-eight and seventy-two hours and five, ten, twenty-four, forty and sixty days.

It was clear from the gross examination of the brain that ferric hydroxide in colloidal suspension was only slightly, if at all, diffusible. The necrotic and hemorrhagic central core of the lesion appeared deep brown, and the zone of reaction, extending for from 3 to 4 mm., light to moderate brown.

*D. Blood Pigment.*—The behavior of the interstitial cells toward blood pigment was investigated by the production of a traumatic cerebral hemorrhage, with consequent formation of hematogenous pigment. Ten rabbits were employed for this purpose. They were killed with chloroform forty-eight and seventy-two hours and five, ten, fifteen, twenty, thirty, forty, sixty and ninety days after operation.

*E. Bacteria.*—The ingestion of bacteria by cells is a true phagocytic process. A strain of small *Staphylococcus aureus* isolated from a carbuncle, a strain of *Streptococcus haemolyticus* (strain 165) from infected tissue of a rabbit and a strain of *Corynebacterium ulcerans* isolated originally by Dr. Ruth Gilbert of the New York State Department of Health from a patient with ulcerative laryngitis were used. The last-named organism was selected because of its short length and because its morphologic characteristics precluded possible confusion with silver granules. All of the organisms were grown aerobically in dextrose brain broth and had a density in barium sulphate of from 1.5 to 2. In twenty rabbits, the cerebrum was exposed, and after injury of the leptomeninges 2 or 3 drops of the culture were allowed to fall from a glass capillary pipet over the injured leptomeninges. With the use of this procedure instead of direct injection of the bacterial culture into the nerve tissue, the micro-organisms were not forced into phagocytes, and the technical error of a false phagocytosis was avoided.

The animals treated with *C. ulcerans* were discarded because of the failure of the micro-organisms to multiply in loco. However, the injections of *Staph. aureus* and *Str. haemolyticus* produced infection in all of the animals, seven in each group. These animals were killed from two weeks to three months after operation, so that both acute and chronic lesions were studied and the time factor was eliminated. The streptococcal infections showed a spreading type of inflammation, with rare minute abscesses in the brain, while the staphylococcal infections were characterized by an occasional epidural abscess and by single or multiple abscesses in the brain walled off by a zone of glial and granulation tissue and associated with suppurative meningitis.

*F. Fats and Lipoids.*—The study of the reaction of the interstitial cells to neutral fats and lipoids presented two difficulties: (1) the inability to distinguish with certainty endogenous fat resulting from cytoplasmic degeneration from phago-

cytosed fat, and (2) the rapid breakdown and disappearance of stained neutral fat or lipoid emulsions. Thus, after the intraperitoneal injection into two rabbits of 10 cc. of a sterile solution of neutral fats of magnesium stained a deep orange color with scarlet red, the fat disappeared within two hours, as observed in frozen sections of the liver impregnated lightly according to the Hortega technic for macrophages.

In order to overcome these difficulties and to have as wide a variety of phagocytic test substances as possible, efforts were made by several chemists to prepare a colloidal particulate suspension of a neutral fat or lipoid which would satisfy all of the following requirements: (1) complete chemical inertia toward all of the reagents employed in the histologic technic to be described later; (2) staining by fat or lipoid stains, and (3) complete suspension of submicronic fat or lipoid particles. Unfortunately, such a preparation could not be developed, and therefore no study was made along these lines.

#### HISTOLOGIC TECHNIC

Immediately after the death of the animal, in order to prevent autolytic changes, especially in the oligodendrocytes, which are highly susceptible, Cajal's fixing solution, freshly prepared, was injected into the carotid artery toward the brain in order to secure the best possible cell preservation. A rectangular block of brain tissue with the wound in the center was then removed and immersed in Cajal's fixative. Many blocks taken at random from both cerebral hemispheres were also cut in order to rule out spontaneous encephalitis or other pathologic conditions. The blocks were frozen on the Sartorius freezing microtome and sectioned in the transverse axis of the wound at a thickness of from 10 to 15 mm.

Reduction of the black, silver-impregnated cell images was carried out to reveal simultaneously the complete cell and the phagocytosed test substances, thus eliminating a prime difficulty accounting for the differences in the results reported in the literature. The term "reduction" is used in its photographic sense, that is, the oxidation of some silver from the image by means of a reducing solution, with the production of a less intense or weaker image.<sup>27</sup>

The following criteria for reducing solutions for use in tissues were established and were completely satisfactory: (1) absence of oxidation of colloidal solutions; (2) uniformity of reduction in proportion to the intensity of the cell image without loss of cell structure; (3) formation of a permanent cell image composed of very fine and crowded lightened silver particles, and (4) ease of control of the degree of reduction.

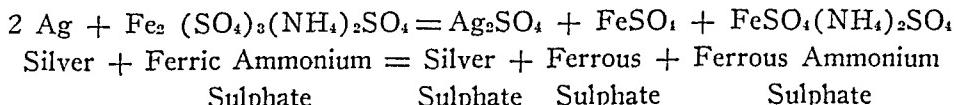
After many trials of various silver reducers, ferric ammonium sulphate was found to meet completely all of these requirements, except in the case of blood pigments, which it oxidized, and it was adopted as the silver reducer for trypan blue, india ink, colloidal ferric hydroxide and bacteria. The original formula of Krauss<sup>28</sup> was modified by doubling the concentration of ferric ammonium sulphate, and the solution was prepared as follows:

Ferric ammonium sulphate (chemically pure) .....	4 Gm.
Sulphuric acid (concentrated) .....	1 cc.
Water to make.....	100 cc.

27. Elementary Photographic Chemistry, Rochester, N. Y., Eastman Kodak Company, 1924, p. 38.

28. Krauss, H.: Ztschr. f. wissenschaftl. Phot. 18:192, 1919.

Ferric ammonium sulphate acts as a proportional reducer, and photographically its reaction, according to Crabtree and Muehler,<sup>29</sup> is as follows:



After immersion for twelve hours at room temperature in this solution, no oxidizing action was exercised on many trial frozen sections of mouse liver deeply stained with trypan blue, india ink, etc., and impregnated according to the Hortega technic for macrophages. Examination of these sections under the Zeiss comparison microscope, in contrast with those not reduced by ferric ammonium sulphate, revealed no loss of particles in the so-called reticulo-endothelial cells. Similar preparations of a kidney from a guinea-pig into which a lethal dose of *Bacillus anthracis* had been injected subcutaneously showed relatively slight reduction of the black-appearing bacilli as contrasted with the more marked reduction of the parenchymatous cells.

The reduction of each section was carried out at room temperature and carefully controlled under the microscope because of the frequent variation in intensity of silver impregnation in various fields of the same section.

The most suitable reducer for blood pigments was found to be potassium dichromate, which is photographically subtractive in action and attacks the photographic silver image, transforming it into silver carbonate and silver sulphates. It does not oxidize the blood pigments. The solution was prepared according to the formula of Crabtree and Muehler, as follows:

Potassium dichromate.....	0.1 Gm.
Sulphuric acid (concentrated).....	0.1 cc.
Water to make.....	100 cc.

For the simultaneous demonstration of microglia cells and trypan blue, india ink, colloidal ferric hydroxide and bacteria, Hortega's technic as described by Penfield<sup>30</sup> was used with a few minor variations. A "strong" silver carbonate solution (molar) was substituted for the "weak" solution, as it yielded more constantly satisfactory results than the latter. After impregnation in the "strong" solution and subsequent reduction in a dilute solution of formaldehyde, U. S. P. (1:100), followed by thorough washing in triple-distilled water, the silver image was reduced with ferric ammonium sulphate for from a few seconds to two minutes, until all the microglia cells appeared from light brown to brownish gray. Toning with gold chloride was omitted, but the cell images were fixed in hypotonic solution and then thoroughly washed in four successive changes of triple-distilled water to wash out completely the residual silver extracted from the cells. The sections were then dehydrated in graded alcohol and cleared in either oil of origanum or a mixture of 95 per cent alcohol and carbol xylene, followed by two changes in xylene, and finally mounted in euparol.<sup>30a</sup> The sections impregnated with bacteria were controlled with the Gram-Weigert stain.

29. Crabtree, J. I., and Muehler, L. E.: J. Soc. Motion Picture Engin. 17: 1001, 1931.

30. Penfield, W., in McClung, C. E.: Handbook of Microscopical Technique, New York, Paul B. Hoeber, Inc., 1929, p. 380.

30a. A mixture of camsal, sandarac, eucalyptol and paraldehyde, used as a mounting medium instead of balsam.

For the simultaneous demonstration of microglia cells and blood pigment, impregnation for microglia cells was practiced as usual by very rapid reduction with potassium dichromate for from five to fifteen seconds, followed by thorough washing in triple-distilled water. Fresh 2 per cent potassium ferrocyanide and 1 per cent hydrochloric acid solutions were warmed to 37 C., and in a few instances to 56 C. The sections were immersed in each solution for five minutes, and the process was completed as usual except for mounting in Gurr's medium in order to preserve indefinitely the reaction to prussian blue. The Cajal fixative used was tested for iron and found to be free from it. The accuracy of the reaction was checked by staining at the same time sections of tissue known to be free from iron.

To demonstrate simultaneously oligodendroglia cells and trypan blue, india ink, colloidal ferric hydroxide, bacteria and blood pigments, the technic for oligodendroglia cells described by Penfield<sup>31</sup> was used, and only slight reduction with ferric ammonium sulphate or with potassium dichromate was found necessary to lighten the cells.

To demonstrate simultaneously astrocytes and trypan blue, india ink, colloidal ferric hydroxide, bacteria and blood pigments, Hortega's regular technic for astrocytes was employed, with ferric ammonium sulphate as a reducer, except that potassium dichromate was used for blood pigments.

In all of the preparations mentioned the general background varied from light yellow to light gray. The interstitial cells appeared from light brownish gray to light gray, and their processes, perinuclear cytoplasm and nuclei were clearly outlined and readily visible. The contrast between the cells and the colored particulate matter and bacteria was sharp.

The bacteria were relatively less reduced by the ferric ammonium sulphate than the cells, appearing from dark brownish black to black in contrast to the light brown to gray color of the cells. Unless involuted, their structure was identical with that shown in the Gram-Weigert preparations, and their recognition offered no difficulty.

#### TERMINOLOGY

The phagocytic cells of the brain have masqueraded under many names. Nissl introduced the term "*Gitterzellen*," now in wide use to indicate the lattice-like structure of the protoplasm after extraction of the fat. His term is objectionable because it indicates only one of many morphologic aspects assumed by these cells, their morphology being determined primarily by the nature of their biologic activity. Merzbacher's designation, "*Abraumzellen*," is more suggestive, calling to mind the removal of tissue débris and foreign bodies. Undoubtedly the name given by Metchnikoff, "macrophages," is the most suitable because of its connotation of the biologic property of the cells, the power of phagocytosis; this name is wisely chosen, moreover, because it does not imply a fixed ancestry. Therefore, the term "macrophages" will be used in this study.

For the classic neuroglia cells, the classification of Andriezen, grouping these cells into protoplasmic and fibrous astrocytes, will be adopted. I shall adhere also to Hortega's descriptive terms, "microglia" and "oligodendroglia."

31. Penfield,<sup>30</sup> p. 378.

## MICROSCOPIC OBSERVATIONS AND INTERPRETATIONS

In control sections of the brains no microscopic evidence of any spontaneous pathologic lesions, such as perivascular mononuclear cell infiltration or focal necroses, was observed. The diffusion of trypan blue was most marked at the margins of the wound and faded gradually so that only scattered dye clumps were present in the normal-appearing nerve tissue. Some of the capillaries and small veins contained many crowded particles and clumps of trypan blue in their lumens. The colloidal ferric hydroxide and india ink preparations showed only a very slight degree of diffusion. The india ink was commonly present as solid irregular masses about the puncture tract and occasionally within the capillaries and small veins, and also, but rarely, at a distance away from the wound as scattered particles and clumps within the veins or the tissues. The ferric hydroxide never showed the clumping and massing manifested by the india ink, and most of the particles were concentrated in the zone of reaction and in the central core.

The sections impregnated with staphylococci revealed intense multiplication of the organisms, which were scattered over a wide area and were present in especially large numbers within the reactive zone. In contrast, the sections impregnated with streptococci showed fewer micro-organisms.

All of the sections were examined with the oil immersion lens. The criteria adopted for injury or death of the cells were those of Lewis and McCoy:<sup>32</sup> (1) loss of color of vital dye granules, (2) diffuse staining of the cytoplasm and nucleus and (3) the presence of particles of vital dye in the nucleus.

The close relation of form and function requires a somewhat detailed description of the structural changes in the interstitial cells reacting to the experimentally produced lesions.

*Microglia.*—Microscopically there was early and sharp mobilization of the microglial cells surviving injury in both the gray and the white matter outside of the limits of necrosis or hemorrhage. It embraced a more or less broad surface, depending on the type of lesion, but was wider and more intense around the experimental abscesses. The microglial reaction was presumably aroused in response to the chemotaxis exercised on the cells by the necrotic and degenerated products set free by the injury or death of the constituents of the neural parenchyma and by the presence of fixed vital dyes, blood pigment and bacteria. The microglia cells were the dominant cells in the acute lesions, rather than the oligodendroglia cells, as described by Cramer and Alpers. In the three day preparations changes of an evolutional rather than of a destructive character could be readily traced from the normal quiescent microglia cell, whether monopolar, bipolar or multipolar, to the fully developed

32. Lewis, W. H., and McCoy, C. C.: Bull. Johns Hopkins Hosp. 33:284, 1922.

macrophage. The microglial reaction was brought about characteristically by mitotic division.

The initial changes appeared to consist of slight retraction and enlargement of the delicate lateral spines and of generalized early swelling of the entire cytoplasm of the cells. At this stage the nuclei retained

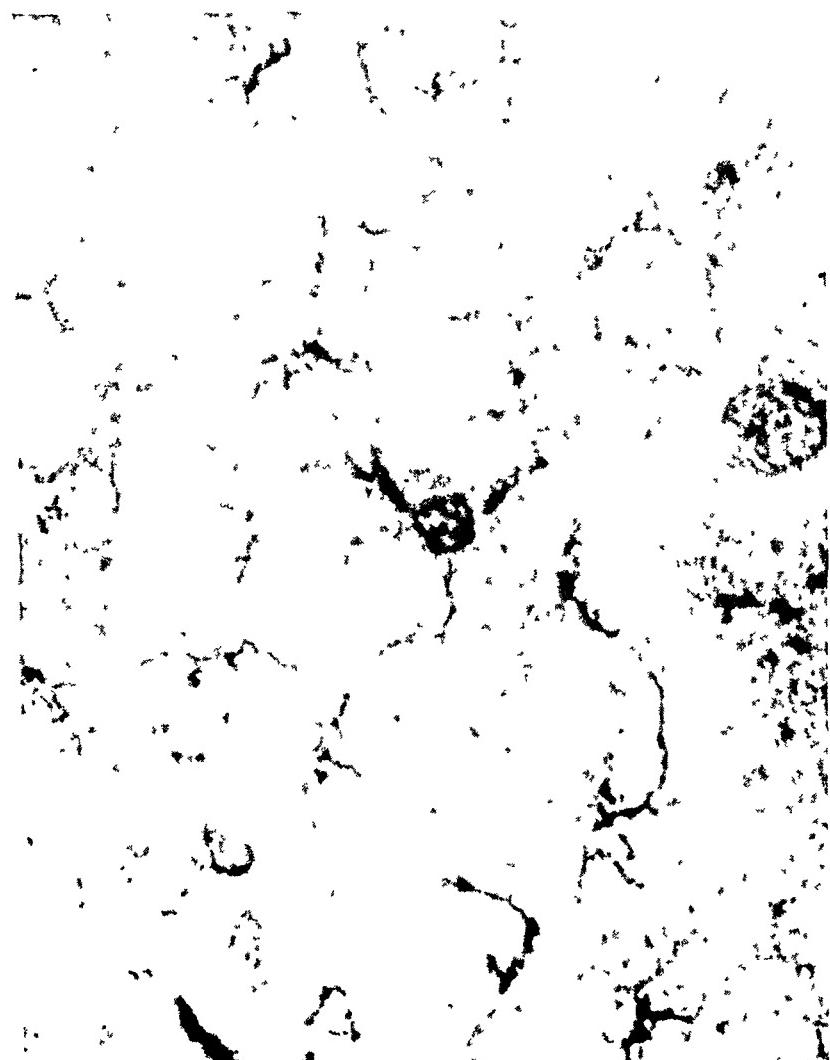


Fig. 1.—A hypertrophied microglia cell (center) containing crowded or scattered trypan blue particles in its thickened, retracted cytoplasmic processes (intracarotid injections of trypan blue; Hortega's silver carbonate technic for microglia cells; reduction with ferric ammonium sulphate; filter H 45 [Eastman]; Wratten "M" plate; magnification,  $\times 1,100$ ).

more or less of their characteristic form, and the cytoplasm was composed of crowded, fine, argentophilic granules.

As the center of the lesion was approached, the cell bodies continued to enlarge and became rounded, with progressive retraction, thickening and loss; first the secondary lateral spines were thus affected and then

the main branches. The nuclei appeared more or less black and more varied in shape. Bipolar rod-shaped cells with long, straight or curved nuclei and elongated, thickened processes streaming out from each end of the perinuclear cytoplasm were not infrequently seen.

The structural changes continued in the next phase, culminating in the formation of macrophages. The outstanding microscopic feature of the cells was their remarkably varied polymorphism, which may have been conditioned by the initial form of the cells; this could not be

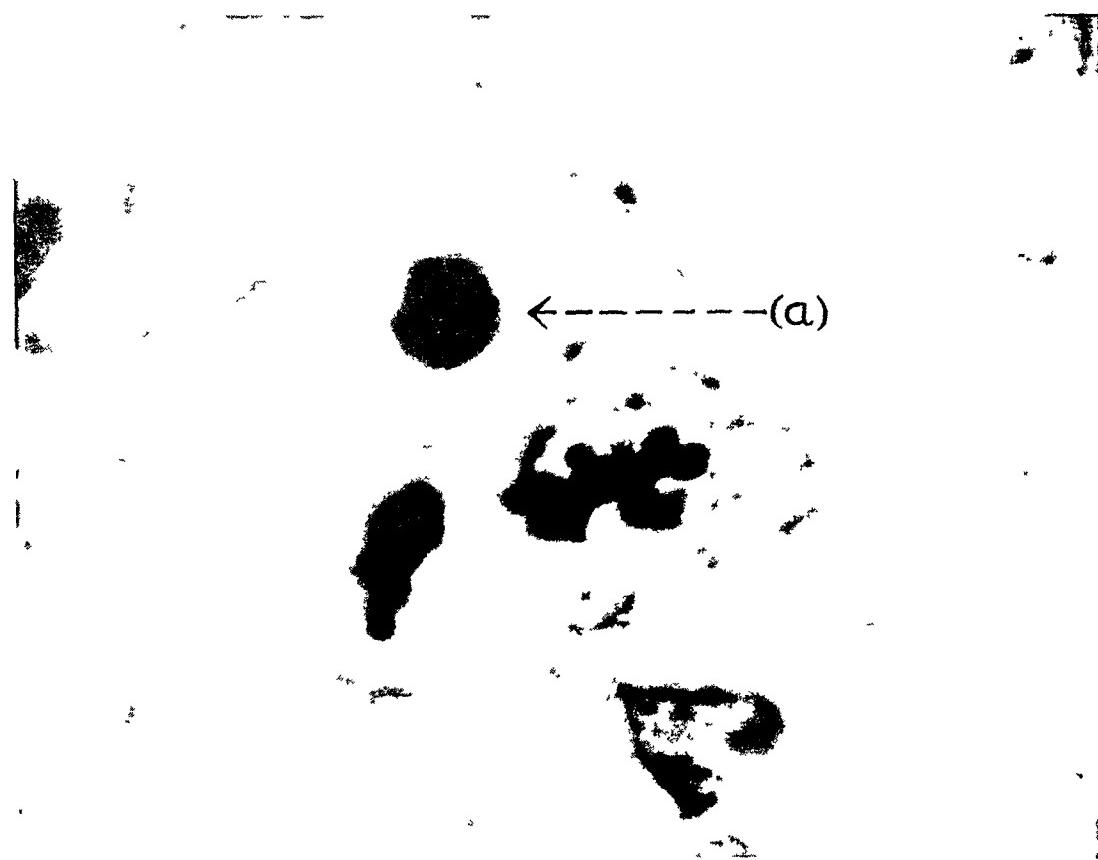


Fig. 2.—Early acute swelling of an oligodendroglia cell (a) lying close to masses and particles of India ink injected intravenously, showing absence of carbon particles in the cytoplasm of the cell (Hortega's silver carbonate technic for oligodendroglia cells; slight reduction with ferric ammonium sulphate; magnification,  $\times 1,800$ ).

verified with certainty, however, by observation of fixed sections. The nuclei were impregnated either darkly or lightly. There were further enlargement and rounding of the cell bodies. Some of the transitional cells with one or two short, rounded or pointed, tuberose stumps resembling pseudopodia were practically indistinguishable from ordinary macrophages fixed in the act of movement. Cells of this type persisted in the subacute and chronic stages but were especially rich in the reactive

zones of the acute experimental inflammations. Their cytoplasm was composed of argentophilic granules of varied form and dimension and presented a more or less well developed spongy, reticulated structure. A relatively small number of the cells had undergone partial to complete cytalysis. Their nuclei were inseparable from the cytoplasm, and they

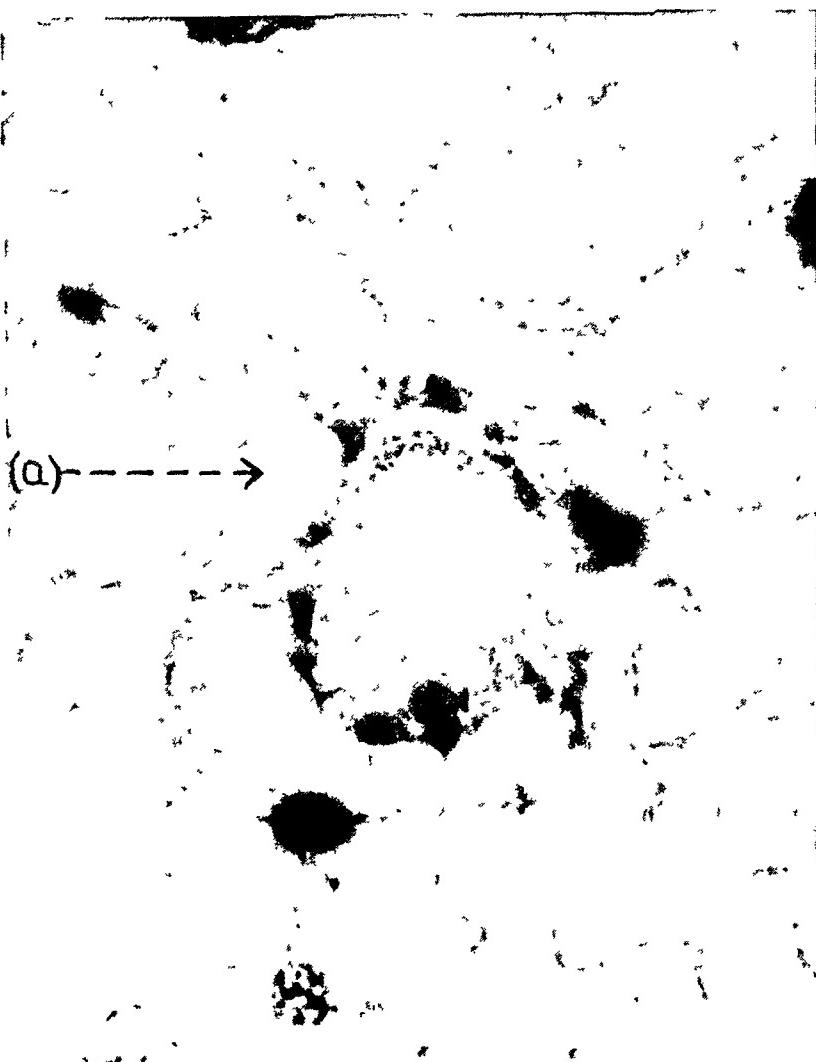


Fig. 3.—A cerebral capillary with india ink particles in the cytoplasm of the endothelial cells, showing absence of carbon particles in the perivasicular foot of a fibrous astrocyte attached to the wall of the capillary (a) (Hortega's silver technic for astrocytes; reduction with ferric ammonium sulphate; magnification,  $\times 1,350$ ).

also showed the usual microscopic changes of injury or death in the vitally stained preparations.

Most of the microglia cells were widely distributed, but some had mobilized as satellites about degenerated or necrosed ganglion cells, nerve fibers and astrocytes. Some lay close to capillaries and small

blood vessels but never applied their cytoplasmic processes to the walls of the former or to the pia-glial membrane surrounding the latter.

A minority of the transitional microglia cells stored a variable number of discrete or crowded particles of trypan blue, colloidal ferric hydroxide, india ink and blood pigment and had phagocytosed as well both staphylococci and streptococci.

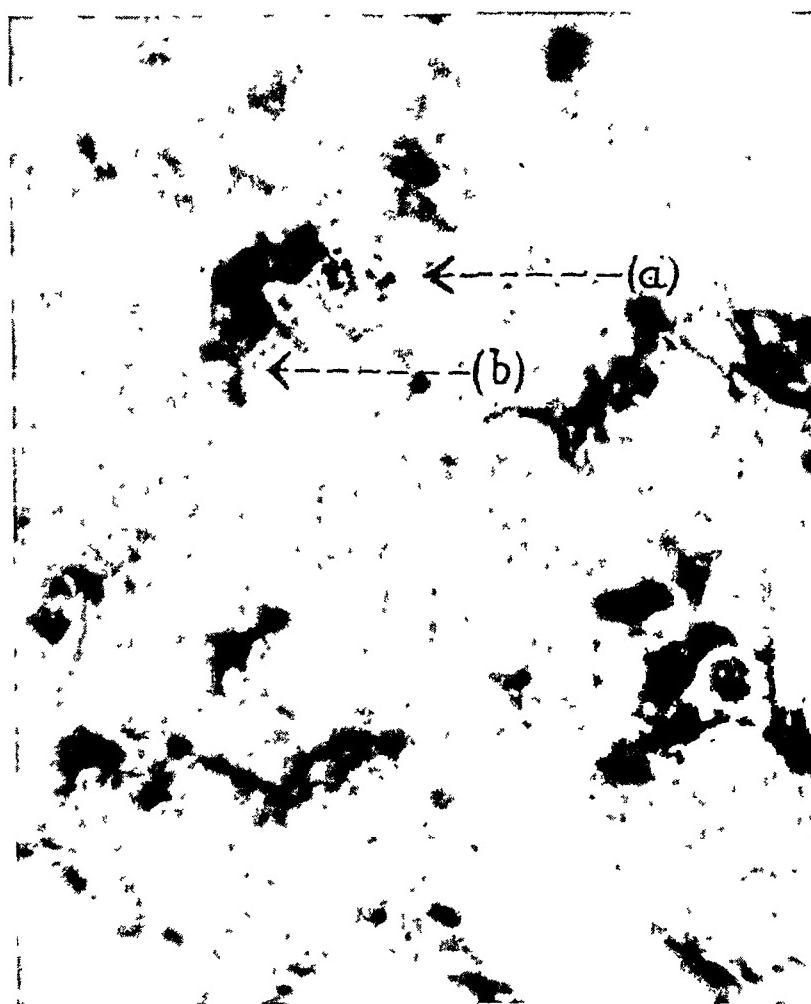


Fig. 4.—Hypertrophied, transitional microglia cells within the reactive zone about an experimentally induced *Staph. aureus* abscess (Hortega's silver technic for microglia cells; no reduction; magnification,  $\times 1,100$ ). This figure illustrates the difficulty of distinguishing bacteria (a) from unreduced microglia cells (b) lying about them.

A few microglia cells with colored particles of varied form and dimensions were noted. These were interpreted as representing the technical error of the phagocytosis of previously stained extracellular material pointed out by Bratianu and Guerriero. Only those cells with uniformly sized and shaped particles of vital dyes in their cytoplasm were accepted as examples of true phagocytosis. Undoubtedly, the

storage of dye particles leads to the development of cytoplasmic changes such as those observed in the transitional cell forms.

It must be emphasized that the majority of transitional cell forms did not store even readily accessible blood pigment granules or vital dyes. In this respect, they behaved differently from specific endothelial cells, which regardless of their situation take up colloidal dye with great avidity. On the other hand, they resemble in a restricted sense normal lymphocytes which never ingest vital dyes until, according to Maximow, they are transformed into monocytes or macrophages. It seems that

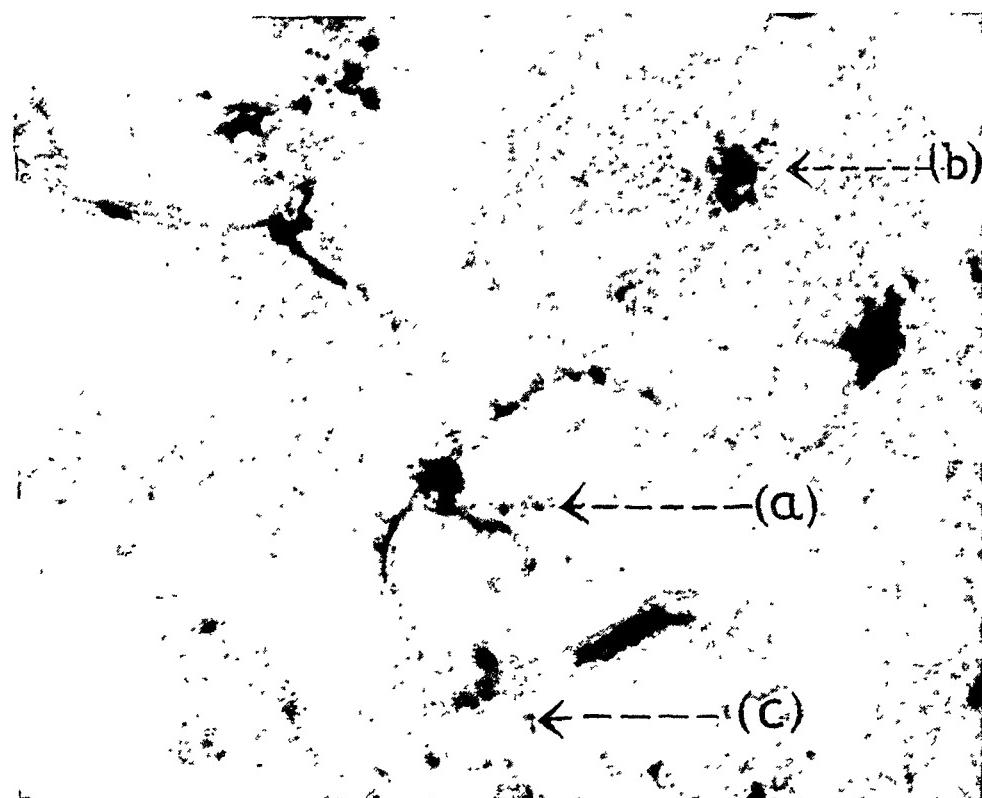


Fig. 5.—Phagocytosis of swollen micrococci, *Staph. aureus* (a), by a hypertrophied microglia cell. Note the neutrophile with phagocytosed micrococci (b) and scattered micrococci (c). (Hortega's silver technic for microglia cells; reduction with ferric ammonium sulphate; magnification,  $\times 1,200$ .)

the microglia cells must be activated by a sufficiently strong stimulus and pass through certain structural changes before they can carry out the function of phagocytosis.

Most of the dye-storing cells were observed among the moderately to far advanced transitional cells and in the animals which were given the largest amounts of vital dye. The nearer the cell approached in structure the macrophage, the more dye it ingested. The persistence of the function of the microglia cells after storage of vital dyes, whatever the mechanism may be, indicates that the structures taking up the

dyes are not vitally concerned in the life of the cell. Only the cells saturated with dye eventually died.

No storage of vital dyes, blood pigment or bacteria was observed in normal or dividing cells. The absence of dye in cells undergoing mitotic division is undoubtedly bound up with the problem of phagocytosis, the exact mechanism of which is still obscure. It is also evident that there

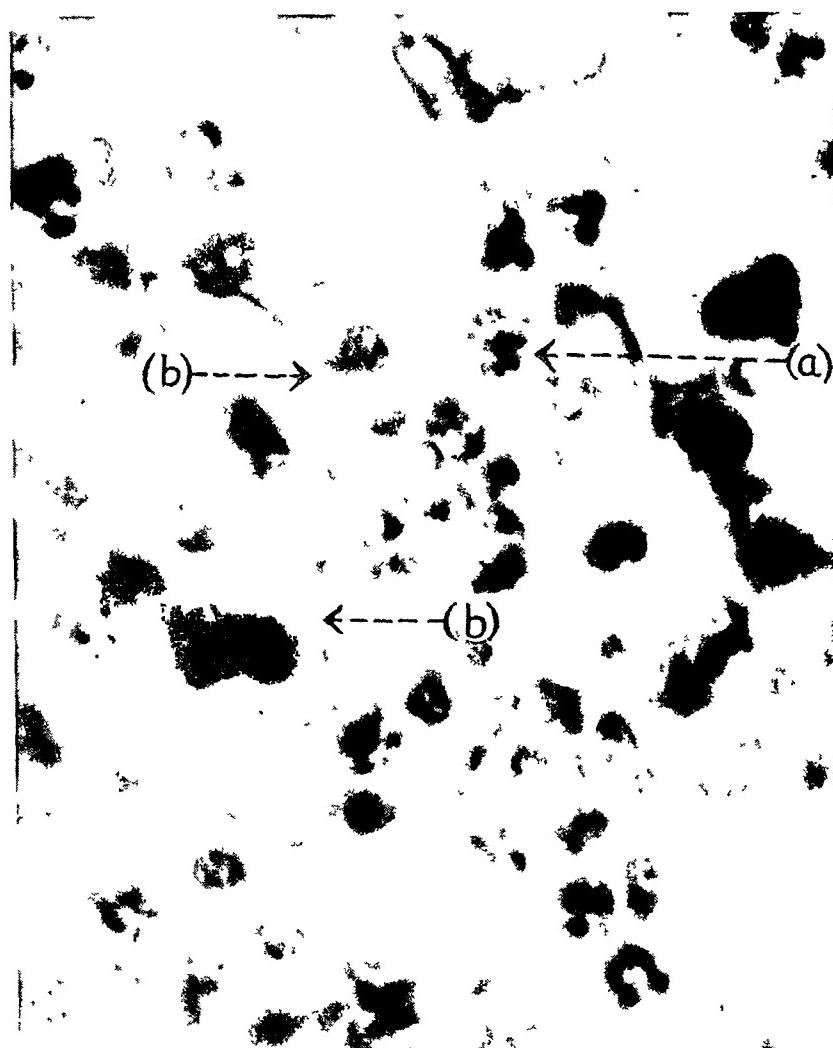


Fig. 6.—Proliferated fibrous astrocytes within an experimentally induced *Str. haemolyticus* inflammation. Note phagocytosis of micrococci by neutrophils (a) and their absence in the neuroglial processes of astrocytes (b). (Hortega's silver technic for astrocytes; reduction with ferric ammonium sulphate; magnification,  $\times 1,350$ .)

exists some relation between the maturity of the microglia cells and their power of phagocytosis for colloidal dyes, bacteria, etc. As for the absence of dye in the normal resting cell, there are two possible explanations: (1) that because of the almost negligible visible quantity of dye

in their neighborhood the cells do not store dye in a sufficient amount to be observed microscopically, because the particles of a vital dye must aggregate in clumps of a certain size before they are microscopically visible and (2) that the microglia cells do not possess the power of locomotion and therefore ingestion will not take place unless the cell and foreign dye particles or bacteria come into physical contact with each other.

The storage of vital dye is not an exclusive property either of macrophages or of cells of mesenchymal origin. *In vivo*, there are several exceptions to the rule that all dye-storing cells are mesenchymal in origin. For example, it is recognized that the epithelial cells of the choroid plexus are of ectodermal origin, and yet these cells store dye in large quantities without furnishing phagocytes. Therefore, although the transitional microglia cells in the rabbit's brain reveal a definite capacity to take up vital dye, a mesenchymal origin cannot be ascribed to them on this basis alone. In a personal communication, Maximow stated: "All macrophages, irrespective of their situation, are exclusively mesenchymal in origin. We can affirm this not because they have the properties of taking up the vital dye—any cell can, after all, ingest dye particles under suitable conditions—but because this is known from innumerable embryological and pathological conditions." Therefore, the decisive demonstration of vital dyes in transitional microglia cells, coupled with the basic investigations of Hortega, Pensfield and others indicating their origin from embryonal meningeal polyblasts and their transformation into macrophages under pathologic conditions during adult life, points to their mesenchymal origin.

The phagocytosis of blood pigment by some of the transitional microglial cells was observed as early as the fifth day after the experimental production of hemorrhage, increasing proportionally with the increase in the age of the hemorrhage.

As in the case of vital dyes and blood pigment, the resting and dividing microglia cells never phagocytose bacteria. This function is performed in the first three or four days of the experimental infection primarily by the polymorphonuclear leukocytes, and only later by a minority of the transitional microglia cells. Thus, the capacity of transitional microglia cells to phagocytose both bacteria and vital dyes runs *pari passu*.

**Neureglia.**—The changes in the astrocytes vary in degree, depending on the nature, severity and duration of the lesion and on the type of astrocyte involved. The most intense regressive and hyperplastic changes were observed about the experimental abscesses. There is a mixed reactive zone composed principally of macroglia and to a lesser extent of granulation tissue about the wound tracts, abscesses and hemorrhage. The earliest reactive changes in the astrocytes were observed in the

forty-eight hour preparations. The astrocytes do not appear to enter into a syncytium but seem to react as separate cellular units. They show slightly increased irregularity, nuclear pallor and fine granular and pale irregular swelling of their cytoplasmic and fibrous processes, associated not infrequently with neutral fat inclusions. The latter are evidently of intracellular origin, since no extracellular fat was observed in the neighborhood of the cells. In the acute stages, the astrocytes situated directly about the center of the lesions frequently undergo partial to complete clastomatodendrosis, as described by Cajal. The cell bodies are markedly swollen, the nuclei show karyorrhexis or pyknosis and the processes are disintegrated or broken up into fragments, clustered about the cellular remains. The great majority of them evidently disappeared, as they were very rare in the older lesions, especially in the traumatic lesions under discussion. In fact, there was a conspicuous absence of astrocytes immediately about the wound tracts and the center of the abscesses in the three and five day preparations.

Astrocytes corresponding to the *gemästete* cells of Nissl were frequent in the margins of the chronic wounds and abscesses. They presented a voluminous, finely granular cytoplasm from which passed forth thick, irregular processes and large, darkly impregnated, eccentrically placed nuclei. The more chronic lesions also showed swollen cytoplasmic bodies and irregular swelling and varicosities of their dendrites in the process of dendrophagia by satellite cells.

Before the third day there was no tendency to increase on the part of the astrocytes, but thereafter they multiplied by amitotic or direct division. The reactive zone of hyperplastic astrocytes was progressively broadened. The damaged nerve tissue appeared to act as a stimulus to the macroglial reaction, for with its gradual disappearance the reacting astrocytes were reduced proportionally in number.

After division, the daughter cells generally took more or less the form of the mother cell, but they differed from the normal astrocyte by possessing more pale, irregular and enlarged nuclei and more rounded cell bodies. Binucleate forms were common. Not infrequently, especially about the abscesses, transition forms, apparently between the protoplasmic and the fibrous astrocytes, were noted. Two weeks after the production of the lesion, the astrocytes were present in conspicuous numbers and threw out fibrous processes to form a young glial feltwork laid down radially about the wound and abscesses. At the end of twenty-four days, the macroglial reaction had reached its height, the glial feltwork showing increased thickening and containing relatively sparse nuclei.

The membrana limitans gliae beneath the pia mater, capillaries and small veins frequently showed regressive changes and contained within

its meshwork occasional wandering macrophages, astrocytes and cocci, single or in clumps, but never any microscopically visible particles or aggregates of colloidal dyes.

Except for occasional diffuse or granular vital staining of dead or injured astrocytes, the latter, whether present in an acute or chronic traumatic or infective lesion, fibrous or protoplasmic, mature or immature, hypertrophic or hyperplastic, preserved or degenerated, under no circumstances stored vital dyes or phagocytosed bacteria or blood pigments.

*Oligodendroglia.*—The initial reaction of the oligodendrocytes in the first seventy-two hours after injury consisted of the characteristic changes described by Penfield and Cone as acute swelling. This reaction was transitory, being rarely encountered in the later stages. It was, as a rule, more regular and intense in the gray matter about the lesion than in the white matter. The cells underwent hypertrophy, with a voluminous increase in cytoplasm and pyknosis of the nuclei, followed by hydropic degeneration of the cytoplasm. A certain number of the cells disintegrated and disappeared, while others reverted to their original state. They were frequently massed about blood vessels, and their short expansions passed occasionally transversely or parallel to the blood vessel wall but never into the pia-glia membrane. Other cells were mobilized as satellites about degenerated ganglion cells.

As early as the third day after injury slight hyperplasia was observed, which increased somewhat one week later but subsided thereafter.

The oligodendrocytes did not appear to give rise to macrophages and never exhibited any phagocytic activity toward colored colloidal ions, blood pigments or bacteria.

*Blood Vessels.*—The local activity of the capillary endothelial cells in the immediate vicinity of the wound tract was slight in contrast to that observed around the abscesses. They rarely showed signs of increase, and mitotic figures were extremely uncommon. Many endothelial cells, especially those situated at the margins of the wound, were swollen as a result of cytoplasmic degeneration or of immaturity. These sometimes contained blood pigment granules, or more rarely particles of trypan blue or colloidal ferric hydroxide, when injured, but no living cells stored either of these two colloidal solutions frequently present in the lumens of the vessels. In the sections impregnated with india ink the common endothelial cells constantly stored carbon particles, but no storage was noted in common fibroblasts. According to Lang, this phenomenon is not a true active phagocytosis but is due to a peculiar condition of the surface tension of the cells. No transformation of any of the endothelial cells into macrophages with desquamation into the lumens of the vessels was observed. Failure of the endothelial cells

to take up colloidal ions or bacteria places them outside the class of phagocytic specific endothelial cells.

Sprouting young capillaries about the wound tract were observed only occasionally and were never sheathed by adventitia and rarely invested by dye-containing phagocytes. Polymorphonuclear leukocytes were present for seventy-two hours, disappearing almost completely thereafter. Careful search within the vessel lumens for cells having the morphologic characteristics of monocytes or macrophages was entirely fruitless. The absence of such cells suggests that in all likelihood no phagocytes reach the brain from other organs.

Fusiform, rounded and enlarged cells containing dye, blood pigment and bacteria were constantly observed within the adventitia of small, medium-sized and large vessels. Comparison with sections taken from rabbits which were neither traumatized nor inoculated with dye showed that these cells could not be distinguished from other cells situated in the adventitial mesenchymal network. Further comparison with sections from inoculated but nontraumatized rabbits showed that in the traumatized brain there was a constant and definite increase of dye-containing cells, which was due to proliferation *in loco* rather than to any cells accidentally wandering into the network. These cells correspond to the adventitial macrophages described by Marchand and Renaud in fixed and stained sections. They represent a barrier to the diffusion of dye into the nerve tissue, supplementing the protective function of the meningochoroidal barrier.

The perivascular spaces which accompany the perforating vessels were dilated and frequently distended with dye-storing or nondye-storing macrophages. Their lining mesothelial cells also stored dye in large quantities. By desquamation and passage into the perineuronal spaces about the ganglion cells, these cells may possibly represent the so-called neuronophagic glial cells, but this could not be demonstrated with certainty in sections cut in one plane.

*Macrophages.*—The structure of the macrophages was varied, depending on the nature of the ingested substances. They were most abundant in the zone directly about the necrotic or hemorrhagic centers. They ranged from 9 to 15 mm. in diameter, the younger forms being, as a rule, smaller than the older ones. When free and fully developed they were large and rounded and possessed, as a rule, a small and eccentric nucleus. The nucleus was bounded by a prominent nuclear membrane against which lay most of the chromatin, scanty and usually concentrated into one or more bodies. The cytoplasm was abundant; it was honeycombed or reticulated, and in the spaces between the delicate cytoplasmic strands were interspersed lipoids, blood pigments, nuclear fragments or dye particles. Sections from the staphylococcic abscesses and diffuse streptococcic inflammations showed frequently macrophages

with phagocytosed, broken-down polymorphonuclear leukocytes and bacteria. Infrequently, the cytoplasm of the macrophages was sharply separated into a pale, vacuolated endoplasm, and an ectoplasm, now broad, now narrow, more deeply stained and either vacuolated or reticulated in appearance. Mitotic figures were present, especially in the later stages, but they were never numerous. Amitosis was occasionally seen. Care was taken not to confuse cells undergoing amitosis with deceptive binucleate forms containing a phagocytosed nucleus. A certain number of macrophages were evidently short-lived, undergoing either injury or death, as indicated by diffuse vital staining of the cells or by the entrance of the dye into their nuclei.

The earliest macrophages rarely stored vital dyes, a fact which can be hardly explained on the assumption of specialization of function in such a lowly differentiated cell. This failure seems to be a question of the amount of dye accessible to the cells rather than of any elective affinity, because with the increase in the amount of dye administered, more and more macrophages accumulated dye particles in increasing quantities. Thus in the ten day preparations, at least one-third of all the macrophages contained dye particles in their cytoplasm. The dye granules varied considerably in size, some reaching a diameter equal to that of the cell nucleus, apparently representing the staining of previously ingested necrosed material. No dye crystals were observed within any macrophages.

#### CONCLUSIONS

1. Silver reduction demonstrated the phagocytosis of colored colloidal particles, blood pigments and bacteria by a minority of the transitional microglia cells in experimental lesions in the rabbit's brain but not in normal resting and dividing microglia cells.
2. The storage of vital dyes by transitional microglia cells is an additional fact pointing to their mesenchymal origin.
3. No phagocytosed vital dye particles, blood pigment granules or bacteria were observed in silver-reduced neuroglia and oligodendroglia cells.
4. There is a distinct relationship between the phagocytic capacity of the microglia cells and their maturity.
5. The failure of the endothelial cells of the cerebral vessels of the rabbit to store vital dyes, blood pigment, bacteria, etc., places them outside the class of specific phagocytic endothelial cells.
6. The actively phagocytic adventitial cells of Marchand supplement the protective function exercised by the hemato-encephalic barrier.

## General Review

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### PATHOLOGY OF UNDULANT FEVER

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While the entire literature of undulant fever has been fairly prolific of recent years and not inconsiderable for half a century, the strictly pathologic portion is limited. There are related aspects, though, that particularly concern the pathologist or other laboratorian. The study has involved him not only in the discernment of postmortem evidences but even more in the recognition of the disease during life, in preventive measures and in the baffling problem of treatment.

By far fewer data appear on the morbid anatomy of the disease than on the bacteriology and serology. Pathologists leave the impression that little is to be said thereon. Much of what is recorded has been only incidental to clinical observation. I cite disproportionately here from such scant notation—having a balanced pathologic picture in view—and allow major consideration to the morbid changes, but it is with the express understanding that this phase of the subject does not find a correspondingly substantial footing in the literature.

Few diseases depend more on the laboratory for a clear definition. Except when the disease is known to prevail through the community, a diagnosis can hardly be established without laboratory test. Encountering the more pronounced form of undulant fever in localities where it is generally prevalent, Marston was able to differentiate it symptomatically from typhoid fever and other acute ailments. He was the first to recognize it as a definite disease. Students of the one epidemic known in the United States (Lake) commented on the extreme difficulty of making a diagnosis from clinical symptoms alone early in the course of an outbreak or in the absence of one. Others (Gilbert and Coleman) have shown how often the disease fails to be recognized, its manifestations being too mild to reach the physician or too indistinct to be diagnosed.

Though it is widely disseminated in this country, the condition was long missed altogether. Craig (1906) first recognized the situation, proving a case to have been contracted in this country and suggesting

the likelihood of many, though they were confused clinically with malaria, typhoid fever, tuberculosis, pneumonia, septicemia, relapsing fever, Hodgkin's disease and rheumatism. This confusion has in no way cleared up since. Recent literature only enlarges the list of confusing conditions: typhus fever and tularemia (Mason, 1931), kala-azar (Loewy), influenza (Clouston), surgical conditions such as appendicitis and cholecystitis (Simpson, 1932), pyonephrosis (Demaree) and an indisposition too indefinite for any clinical impression except myocarditis (Clark). Members of the medical profession have diagnosed cases through laboratory procedure.

More than one condition comes into consideration as undulant fever. These various manifestations are caused by distinct varieties of the infective organism and are not identical clinically. The best studied, and until recent years the only one known, is that of caprine origin, which caused devastations at Mediterranean posts compelling the attention of British military surgeons late in the nineteenth century. Earlier literature designates it by any of a dozen or more different names, usually as "Malta" or "Mediterranean fever." Most of the names seemed less and less appropriate as knowledge of the condition broadened, and the name "undulant fever" occurred to Hughes (1896) as a more descriptive and less ambiguous term. Preference eventually settled on this designation, and international endorsement was accorded it in medical gatherings (Blanchard, Bassett-Smith, 1914). Forms of the disease more recently defined, and related etiologically to contagious abortion in the cow and hog, are sometimes referred to in contradistinction as "abortus fever," but more generally they are also designated as "undulant fever."

#### BACTERIOLOGY

The septicemic character stands out in the pathologic picture of the condition. On microscopic section Bruce (1887) found coccoid bacterial forms in the spleen of a patient who died late in 1886, and the following year he cultivated the organism. Later he succeeded in cultivating it from the blood of patients during life. Tissues throughout the body yield the culture: blood from the heart, the spleen, liver, gall-bladder, kidneys, suprarenal glands, pancreas, thyroid gland and many of the lymph nodes (Bassett-Smith, 1922). The organism may remain cultivable for long periods in the blood or other tissue of an infected person. Burnet (1922) found that it was cultivable from bone marrow when not from the blood. A positive culture has been reported from the spleen after eighteen months (Bassett-Smith, 1922) and one from an ovarian cyst after six years (Wainwright, 1929a). Various observers have found the organism in the joints, the tonsils, the ovaries, the oviduct, the epididymus and practically all the parenchymatous organs

(Carpenter and Boak, 1931). Strains vary greatly, however, in the activity with which they invade these tissues.

The micro-organism was described in some detail by Eyre (1907). Though so persistently spherical as to support a definition of coccus (Eyre, 1926), its capacity for rod formation also establishes it in practically all the current classifications as a bacillus. That of Winslow and his collaborators included it in the genus *Bacterium* with a note that data not yet on hand might constitute it a new genus. Meyer and Shaw, and Feusier and Meyer applied to it the name *Brucella*, which ever since has met with general enthusiasm among workers with the organism. The classification of Lehmann and Neumann left it in the genus *Bacterium* but recognized a subgenus *Brucella*, as it did a subgenus, *Pasteurella*. Bergey and his collaborators first classified it in Castellani and Chalmers' genus *Alcaligenes*, attributing its initial omission from the genus to a doubt as to its bacillary form. This generic terminology likewise found proponents among the writers on undulant fever, but in its last revision Bergey's (1933) classification recognizes instead the genus *Brucella*.

*Differentiation of Organisms.*—Literature on bacterial characteristics concerns principally the differentiation between types. Undulant fever was first attributed to the type infective primarily for goats. Evans (1918a) pointed out a close antigenic relationship between it and the bacillus of infectious abortion in cattle. Agglutination tests that readily differentiated the latter from the *bronchisepticus* organism failed to distinguish it from that of undulant fever except by a difference in titer. Further study brought forth many antigenic types. By absorption of agglutinin Evans (1925) distinguished in her series an *abortus*, a *melitensis* A, a smaller *melitensis* B, a *para-melitensis*, a *para-abortus* and three other types of a single strain each. Feusier and Meyer described four serologic groups, with *abortus* strains falling in group I, *melitensis* strains in I, II or III, and *para-melitensis* strains in group IV. Serologic grouping was not a simple one.

Type differentiation was undertaken in a number of laboratories, but far from clearing the situation it disclosed more and more divergence and multiplicity of types. It was suggested that the *para-melitensis-abortus* groups be considered mutant or rough types of the smooth *melitensis-abortus* (Ross, 1927a). Provision for smooth and rough variants leaves the classification even more cumbersome, but the failure to distinguish between them has been held accountable for some of the difficulties (Wilson, 1931). Serologic grouping into so many varieties seemed to Bassett-Smith (1925) premature and confusing, and he regarded the indication of pathogenicity as a more useful guide. Others expressed themselves similarly (Duncan). Not only did the patho-

genicity test avoid what seemed a superfluity of bovine and caprine types, but it also differentiated an important type obtained from swine.

Distinction by pathogenicity rests primarily on the natural infectivity for goats, cattle or swine. Pathogenic activity in laboratory animals helps a little to classify organisms isolated in human undulant fever. Theobald Smith's (1926) descriptions show how strains of the different origins may produce a more or less distinctive pathologic effect in guinea-pigs. One group tends to greater pathogenic activity than another, and strains less highly virulent cause lesser degrees of endothelial proliferation and other morbid change presently to be detailed. The method's weakness in differentiative ability is shown by the variability of result from diverse sources of experimentation (Strong). The most practicable evidence it gives is that porcine strains, indistinguishable serologically from the bovine, produce pathologic effects more closely approximating those produced by the caprine strains. A serologic in conjunction with a pathologic test thus provides some means for separating all the strains into the three groups. Rainsford subgroups his strains serologically and by the production of hydrogen sulphide into melitensis and abortus, and then the latter by inoculation of animals into bovine and porcine strains. He gets better results with hamsters than with the more generally utilized guinea-pigs.

Biochemical behavior offers additional points of distinction between the organism's three main varieties. The differentiation by the hydrogen sulphide test, just referred to, agrees uniformly with that by absorption of agglutinin. This substance is liberated by both of the abortus forms but not by the melitensis, when grown on Stafseth's liver infusion medium, and is indicated by lead acetate (Huddleson and Abell, 1927). The liver medium is often adopted for isolation of the organism. Huddleson, Halsey and Torrey (1927) use it with gentian violet to inhibit the growth of any gram-positive flora present. They cultivate at 37 C. aerobically for fifteen hours, then continue some cultures for ten days aerobically and others for seventy-two hours in an atmosphere replaced by from 5 to 10 per cent carbon dioxide.

The change in cultivation atmosphere facilitates the growth of the bovine type of organism. When first describing his abortus bacillus, Bang suggested a procedure for the reduction of oxygen, though not its elimination by pyrogallol (Bang). Huddleson showed that the advantage of sealed tube growth lay not in the low oxygen tension but rather in the increased concentration of carbon dioxide. Caprine strains, on the other hand, though facultative anaerobes (Eyre, 1907), are much retarded if deprived of a free supply of oxygen and do not benefit by additional carbon dioxide in the atmosphere. The porcine strain, like the caprine, grows in the air from the first generation, and on passage through animals may be reisolated from the tissue strictly aerobically.

(Good and Smith). With fresh strains this same differentiation is made from the degree of utilization of dextrose: not over 2 per cent by the bovine as compared to from 5 to 20 per cent by the other strains (McAlpine and Slanetz).

Huddleson (1931) also described a differential test depending on the varying bacteriostatic action of such dyes as thionine and either methyl violet or basic fuchsin. Thionine in the employed concentration inhibits the bovine, the other two dyes the porcine, and none of them the caprine, strains. In a large illustrative series tested, 325 strains fell into the first, 172 into the second, and 133 into the third, of these respective groups. Unlike the absorption or other tests already described, this reaction by itself differentiates all three types. Its dependability is variously evaluated by others. According to one comparison of a classification of 19 strains on this basis with that by absorption of agglutinin, there was agreement in 9 and disagreement in 10 strains (Francis, 1931).

Recent classifiers of strains have preferred to employ several of or all these various bases of differentiation rather than to choose between them. Serologic, pathogenic or cultural distinction is not of itself sufficiently clear to make it possible to disregard the others. Blake and Oard, for instance, employ and advise absorption of agglutinin, inoculation of guinea-pigs, the carbon dioxide requirement and utilization of dextrose for the routine differentiation of all the human strains.

Classification is carried at least to caprine, bovine or porcine designation. These varieties are generally recognized. Others may assume importance locally, for instance the one prevalent in Rhodesia according to Bevan (1925) and Duncan. Duncan defined this strain as presumably bovine but differing from the bovine as heretofore described in that its cultivation atmosphere need not have additional carbon dioxide. Evans (1925) regards all these varieties as of a single species, *Brucella melitensis*. Others continue to separate as *Brucella abortus* the organisms derived from contagious abortion of cows or swine. Eyre (1926) could not justify from his strains a relationship even this close between the two. Bruce's organism, unlike Bang's, remained persistently coccus-like, and cultural differences seemed distinct.

*Transmission of Infection.*—For many years following Bruce's description of the organism the infection continued as an ill-comprehended scourge disseminated no one knew how. Its devastations were felt continually by British army posts of the Mediterranean and by neighboring civil populations. Eventually the admiralty and war office, cooperating with the civil government of Malta, established the Mediterranean Fever Commission to investigate the cause of the disease and consider preventive measures. Studies conducted from 1904 to 1906 established several points. The infection was septicemic in goats and

sometimes localized in the udder. The common means of contraction by man was through ingestion of milk from infected animals. Less common was subcutaneous inoculation through abrasions incidental to handling (Eyre, McNaught, Kennedy and Zammit). How closely the prevalence in the military and civil population of Malta was related to the consumption of goat milk is brought out by the Commission (McCulloch, Weir and Clayton) and further elaborated on by Eyre (1912).

The likelihood of transmission through goats excited immediate attention elsewhere, and brought contributory observations pro and con. Sargent, Gillot and Lemaire, for instance, determined by agglutinative reaction of the milk and by cultural study the proportion of infected goats in Algiers. Some other potential sources of infection were hard to set aside, and direct contact was still suggested to Ross (1906) by the high prevalence in hospitals, houses or barracks where the disease localized. He observed, too, that sailors picked up the disease ashore and that they did not drink milk.

Undulant fever of the type under investigation at Malta must have prevailed then and ever since in the southwestern part of the United States. Gentry and Ferenbaugh found it endemic through the older goat-raising sections of Texas and noted that a disease of like symptomatology had been known among the people for at least twenty-five years. Thirty-four per cent of the goats tested gave positive agglutination reactions. Yount and Looney associated the occurrence also with the goat industry in Arizona, and the same situation extended to other neighboring states (Kampmeier). Herds of goats thereabouts and in Mexico proved on survey to be extensively infected (Holt and Reynolds). In this endemic area, as in Malta, the control of the disease has been effected primarily and principally through supervision of the supply of goat milk (Tappau).

The undulant fever of caprine origin was encountered and recognized over a large part of the globe before any other form was suspected. In 1914 Bassett-Smith stated that wherever the disease is found, in Italy, India, South Africa and America, goats are practically always present and distribute it, but that other ruminants may carry it. At about that time certain analogies with contagious abortion in cows began to appear. The abortus organism described by Bang had been recognized in this country as elsewhere (MacNeal and Kerr), and Evans (1918b) noted its close resemblance to the organism of undulant fever. She found it present, though not continuously in large numbers, in milk taken from the udders of cows that had aborted, indicating a possibility of human exposure. Other suggestive data accumulated, and Keefer reported a case of undulant fever with evidence of such bovine origin. Subsequent evidence clearly established this etiologic relationship. In

one experiment, pregnant heifers were inoculated with organisms isolated from human cases, and they aborted and yielded the organism in culture from the fetus, placenta and colostrum (Carpenter, 1927).

Histories of patients with undulant fever soon proved to accord with a possible infection from this source. In one series of 38 cases, 17 patients had been in contact with cows that had aborted and 12 drank raw milk (Weigmann). Again, of 82 persons whose occupation brought them into close contact with infected cattle, the serums of 22 gave positive agglutination or complement-fixation reactions or both (Kristensen, Helms and Martensson). Six gave positive agglutination reactions at 1:100 or over, and 1 had a symptomatic case of undulant fever, while control persons all gave negative reactions.

While the abortus organism thus showed a definitely invasive property, this activity was of less degree than that of the melitensis organism. A lesser invasiveness for monkeys on experimental feeding was shown by Fleischner, Vecki, Shaw and Meyer. Burnet (1928) showed the same for man on injection. Morales-Otero's (1929, 1933) feeding experiments in man indicated a less infective character of strains from cow's milk, the strictly bovine in particular. Comparisons drawn by Kampmeier showed that in man such infections ran a milder course clinically than those transmitted by goats.

An extremely low rate of attacks contrasts with the frequency of abortus organisms reaching the milk supply. Carpenter and Boak (1928) found that the cream of over 6 per cent of an arbitrary series of cows' milks contained the organism infective for guinea-pigs. McAlpine and Mickle presented presumptive evidence of wide exposure to infection in Connecticut, and yet contraction of the disease by man was limited in that locality. With 90 per cent of dairy herds infected and only 60 per cent of the milk pasteurized, only 0.6 per cent of 10,157 random serums received for a Wassermann test agglutinated the abortus bacillus in dilutions up to 1:100.

In view of the wide dissemination in market milks, it must be this relative avirulence of the abortus organism that spares much of the world from ravages such as those experienced in Malta. It is interesting to contrast the situation regarding undulant fever about the Mediterranean and other foci of caprine infection with that in countries in which cattle constitute the sole source of infection. There is reason to presume that infected cow's milk has been the only common vector in Great Britain (Dalrymple-Champneys, 1931). The Board of Agriculture and Fisheries long ago confirmed the widespread existence of epizootic abortion among the cattle in England, Wales and Scotland (McFadyean and Stockman, 1909), and the milk or serum of cows in London has agglutinated the organism in high dilution (Kennedy, 1914). The infection of goats does not appear, though Broadbent reported as an exception an atypical case supposedly contracted from a goat.

Great Britain has little clinical undulant fever but apparently a more considerable prevalence of subclinical infection—reasonably attributable to heavy exposure from contact or from the milk of aborting cattle—with low susceptibility (Wade). Cruickshank and Barbour found that a hospital population altogether without evident undulant fever gave positive agglutination reactions in a little less than 0.5 per cent of instances, and they associated these instances with exposure to infected cattle. The same situation was reported for Scotland by Marr and by A. Thompson for Ireland: Infectious abortion of cattle is rampant, but undulant fever in man is exceedingly rare.

It was not long before *Brucella abortus* of the swine type was found among the organisms accountable for undulant fever. The type of *abortus* organism that causes the larger portion of infections in swine had been described (Cotton) as an aberrant type, infective for both cattle and swine, and producing in guinea-pigs a lesion different from that caused by strains affecting cattle alone. Smith (1929) thought that it may have developed from the bovine type through the feeding of hogs with dairy by-products. He found (1926) that strains of this variety produced lesions in guinea-pigs much more like those produced by some human strains than did the bovine variety. Evans (1925) suggested the possibility of porcine infection. The studies made in Iowa particularly showed its wide extent. An instance occurred with 3 infections of the bovine type but 1 strongly suggestive of the porcine type (Awe and Palmer). Further instances accumulated, and the probable proportion of cases of porcine infections increased (Bierring). Kern mentioned infection with undulant fever resulting directly from hog carcasses, but it also developed that many infections from cow's milk were caused by porcine organisms infecting cattle (Blumer). A study of 11 cases of undulant fever in Georgia (Atwood and Hasseltine) indicated their causation by porcine strains transmitted through the milk supply.

The porcine strains show greater virulence for man than the true bovine strains. By feeding them to man, Morales-Otero (1929) produced typical undulant fever with a positive blood culture and in 1 case an agglutination reaction, but he failed with bovine strains. Huddleson (1929) similarly obtained greater pathogenicity for monkeys by feeding. The extent to which contagious abortion gives rise to clinical undulant fever seems to depend on the preponderance of organisms of the porcine type. The infrequency in northern Europe and, as rule, in this country may be attributed to a relative rarity of exposure to porcine strains, to whose attack human tissue is more susceptible (Hasseltine, 1929).

The widespread presence of Bang's bacillus had suggested appropriate treatment of the milk supply even before milk-borne undulant fever was known. Melvin considered contamination with *abortus* organisms another link in the chain of facts establishing proper pasteurization

of all market milk as a measure essentially necessary for protection of the public health. Without professing to know what effect this organism might have on human beings, he would not assume that there was no effect. Recognition of the transmission of undulant fever through milk amply sustained the contention. State health departments have included infectious abortion with tuberculosis as diseases to be eliminated from dairy herds (King). Experiments conducted under the Bureau of Animal Industry (Cotton, 1924) showed that vaccination is futile and suggested more likely control through separation of herds to weed out the infection. Until this can be accomplished, for a number of years anyway, Hasseltine (1930) suggested pasteurization as the sheet anchor in the prevention of milk-borne undulant fever.

Virulent porcine as well as bovine strains in milk are destroyed by pasteurization, though their thermal death point is twenty as compared with fifteen minutes at 140 F. (Carpenter and Boak, 1933). Pasteurization had been early suggested in connection with infected goats' milk, including that used in ice-cream or native cheeses (Eyre, McNaught, Kennedy and Zammit). The organism survives the handling procedures incidental to the manufacturing of these products, and there is a possibility of contracting undulant fever from ice-cream frozen a month or more (R. Thompson).

It soon developed that the disease, even that caused by the strictly bovine strain, is commonly transmitted otherwise than by cow's milk. The transmission appears to be principally by milk in some localities and, according to some, throughout the country (Simpson, 1930). Starr and Maxcy noted that about 60 per cent of the cases in Virginia are so transmitted, compared with 40 per cent of cases due to contact with animals. The extensive incidence in Iowa was not distributed like a milk-borne infection (Hardy, 1928). Milk would probably have brought the largest percentage to the city, whereas 145 of 209 cases were rural. The much greater rate of attack in males than in females and to some extent the vocational and age incidence suggested rather the contact with cattle and hogs (Bierring). Raw meat sometimes appeared to be a disseminating agent (Moorehead).

Contraction of the infection is rare except by transmission in some manner from one of the three kinds of animals. There is no evidence of transmission from man to man (Simpson, 1930). In spite of this a proper disposal of the urine and feces has been urged (Carpenter and Boak, 1933). The organism readily reaches the urine and the feces, apparently through the bile (Bassett-Smith, 1922). Elimination is principally by these excretions.

The danger of accidental infection in the laboratory is by no means negligible and has been said to occur even more readily than with glanders or plague bacilli (Widal, Léon-Kindberg and Coton). While such

cases are rarely reported, laboratory workers have been known to pick up the infection from suspensions or cultures. Huddleson (1926) mentioned an infection of a graduate student working with the organism, though he was more suspicious of raw cow's milk which had been consumed. Before the Mediterranean Fever Commission had demonstrated the common avenue of transmission, accidental infection had already been observed repeatedly. Bassett-Smith (1904) in his description of the undulant fever situation among naval forces at Haslar expressed the belief that the usual path of transmission was by dust and wind, but he pointed out how readily the organism in laboratory cultures caused attacks if taken in by mouth or accidental inoculation. Prior to that he (1902) had mentioned a number of instances of accidental infection.

*Incidence of the Disease.*—Before proceeding with the pathologic identification of cases of undulant fever, one might consider just where and among what groups of persons these cases are likely to be encountered. Distribution charts changed radically, of course, with recognition of the relatively mild forms associated with contagious abortion. They now show the occurrence of the disease practically throughout the civilized world (Dalrymple-Champneys, 1933). When the only known areas of infection showed the goat-borne fever, the condition was passed over with other tropical diseases by most pathologists. The known incidence extended broadly between the forty-fifth parallels north and south through all the continents except Australia, but large areas within this zone seemed unaffected. Apparent freedom from the disease was often due to inadequate diagnostic skill, but some countries with such advanced medical facilities as Great Britain reported no cases (Bassett-Smith, 1922). In the United States practically all the diagnosed cases were imported prior to the reports cited from Texas. Some occurred among persons from abroad, especially soldiers and sailors from the Philippines or other tropical stations (Curry; Craig, 1903; Mason, 1903).

Craig (1906) first reported a case as having been contracted in the United States, that of a nurse in Washington, D.C. Even at that time he thought that the disease was much more widespread than is usually supposed and suggested serodiagnosis of undetermined fevers in all regions. Gentry and Ferenbaugh (1911) described a considerable prevalence of undulant fever in the Rio Grande valley and subsequently a report came of the one epidemic recorded for this country, in Phoenix, Ariz. (Lake). This form of the disease, contracted from goats and due to the melitensis type of organism, has remained limited to that section of the country—Texas, Arizona, Utah and Nevada. The disease so widely described elsewhere is that contracted from cattle or swine and due to the abortus types of organism (Wainwright, 1929a).

Undulant fever due to the abortus organism was supposed to be infrequent when it was first described (Keefer), but during the few

years following, case after case appeared. Unlike the cases of caprine origin, these were generally distributed through the country. A review of the first 20 showed 1 case in Washington, D. C., 2 in Maryland, 1 in South Dakota, 1 in Connecticut, 7 in New York, 1 in California, 3 in Utah, 1 in Virginia and 3 in Michigan (Evans, 1927). The prevalence became most extensive in Iowa (Hardy, 1928). A survey soon showed a greater or less occurrence in three fourths of all the states (Blumer), with reports still coming in from others (Carey and Newsom). It was realized that undulant fever probably exists in all and is likely to prove rather prevalent wherever carefully searched for (Moorehead).

The season exerts some rather inconstant influence on the prevalence. Like other milk-borne infections, undulant fever from the first has been encountered most in the summer (Hughes, 1897). The abortus like the melitensis form has this seasonal tendency. Official tabulations in the United States for 1929 and 1930 showed the larger proportion of cases reported in the summer and autumn, reaching peaks in July and September, respectively, but the duration of the cases prior to report was unknown (Hasseltine, 1931). Scattered observation of others lacks uniformity and fails to attach significance to any seasonal rise noted.

The rural cases far outnumber the urban. The melitensis form presents only one exception, the outbreak in Phoenix (Lake). The clearest evidence in the abortus form comes from the numerous cases in Iowa, with a rate per hundred thousand of 11.4 in rural districts, 8.3 in towns of under 5,000 population, and 4.0 in larger cities (Hardy, 1929c). Reports from abroad, from Denmark for instance (Kristensen, 1928), also suggest a predominance in rural districts and insignificant seasonal changes.

Occupation has more to do with infection from goats than from cattle and swine. Of the 5 cases reported by Ferenbaugh, 4 were in goat herders and 1 in a goat ranchman, and except at Phoenix this occupational relationship is borne out by subsequent observation from that section. Similar influence might be expected with infections from cattle or swine in which milk is not the vector, since proximity to these animals depends on occupation, but it has not been found so distinct a factor (Wainwright, 1929b). In four selected localities during the prevalence of the disease in Iowa, the percentage distribution was as follows: among farmers and their wives, 31.6; employees in packing houses, butchers and veterinarians, 6.9; business and professional men, students and laborers, 39.4; housewives, 16.8; children and invalids, 3.9, and laboratory workers, 1.4 (Hardy, 1929c).

Evidence presumptive of subclinical infections, consisting in a positive agglutination reaction of the blood serum, is sometimes more suggestive of occupational influence. Of 49 veterinarians, 3 of whom had a history suggesting clinical undulant fever, 57 per cent showed agglu-

tinins in the blood and 26 per cent showed a reaction at 1:100 or above (Huddleson and Johnson, 1930). Again, complete agglutination was demonstrated in 33.4 per cent of 120 veterinarians, in 24 per cent of 220 employees in packing houses (13.6 per cent reacted at 1:80 or above) and in 16.5 per cent of 138 consumers of raw milk (3.6 per cent reacted at 1:80 or above) (Jordan).

The age of attack ranges from early childhood to old age, but most cases fall in the 20 to 44 year groups. About two thirds of the patients in the Iowa series were in these age groups (Hardy, 1929c). The infection is exceptional in infancy or early childhood, but cases are described; one occurred in a child aged 1 year on a cow's milk formula (Kohlbry), and 1 in a child of 7 months on breast milk (Hill and Monger). Certain authors in describing the disease in children (Anderson and Pohl) have noted the relatively mild character of these cases, the attack being hardly noticeable except for a persistent fever. They think that the disease may be more frequent in children than is commonly believed.

#### MORBID ANATOMY

The literature is not yet adequate for a clear conception of the morbid anatomy of undulant fever. Authors of textbooks on pathology make little attempt to cover the subject and have practically omitted it except in the most recent issues. Boyd devoted two pages thereto, but only a small part of this matter described pathologic lesions and it featured the toxic and other changes common in septicemias. He mentioned the rarity of postmortem studies in explanation.

The postmortem material is limited by a low fatality rate, less than 2 per cent according to Boyd. Occasional virulent outbreaks have been attended by a considerable number of fatalities. According to the report of one exceptional outbreak 160 of 630 inhabitants contracted the disease, and 40 died (Aubert, Cantaloube and Thibault). The early figures from Malta indicated a fatality rate of 2.3 per cent among 1,705 navy and 1,947 army men, or of 6.9 per cent of a total of 4,627, combining these cases with those in civil administration (Eyre, 1908). The greater proportion of cases of bovine origin are mild and rarely fatal (Leavell, Poston and Amoss). They may have a fatality rate approximating 2 per cent (Kristensen, 1928). With so few of these cases on record as yet, the total rate for undulant fever can only be surmised.

The bulk of the postmortem observations that have reached the literature are practically negative. They rarely establish a postmortem diagnosis, the simplest procedure for which is through culture of the spleen, liver, kidney and mesenteric gland (Topley and Wilson *a*). Morbid changes that would create a picture in any way distinctive have been encountered too rarely to suggest that they are uniformly produced by the disease. On the other hand, since most reports are recent, it is

conceivable that newer refinements of observation and technical procedure are developing a pathologic concept missed until now. The few observations of that nature on record may prove a nucleus for a more characteristic entity.

Earlier postmortem observations are best expressed in a treatise by Hughes (1897). A summarized report is made of 60 postmortem examinations of hospitalized soldiers at Malta; 15 of these were seen by the author. In the acute cases gross examination revealed an intense congestion, especially marked in the internal organs. The chronic cases showed a similar change, though less constant or pronounced, and also various effects of the long continued irritation of the tissues. The microscopic study added nothing of significance except that in the spleen a larger proportion than usual of lymphoid tissue was evident. A postmortem examination of later cases shows little more for the most part, when made as a routine and reported only incidentally as these were. One random report (Hardy, Jordan, Borts and Hardy) gives as the most striking feature a complete absence of any gross pathologic changes, and the only microscopic ones a chronic interstitial pancreatitis, chronic cholecystitis, fatty infiltration and passive congestion of the liver, and myocardial degeneration with fragmentation, not especially significant of undulant fever.

Other recent observations are reported because they attract special interest in the pathologic changes and are perhaps new to the literature. It is these that one scans most hopefully for any characteristic change that may hitherto have escaped notice. One finds them scattered thinly through a volume of inconsequential observations. Unfortunately, they are not only exceptional but highly variable and conflicting. As Wohllwill said in reference to abortus disease in man, there are submitted at the present time quite striking dissimilarities of the pathologic anatomic conditions. Each observation must stand more or less by itself. Since the first report of any extraordinary occurrence bears various interpretation, significant evaluation of these observations cannot immediately be made.

A question arises not only as to the characteristic cellular reactions but also as to the extent to which the infection actually causes the pathologic changes encountered. The uncertainty subsides only as the picture recurs. One finds endocardial involvement first regarded as probably rheumatic (Hughes, 1897; Scott and Saphir), since the literature on undulant fever did not feature it, but later considered as localization of the undulant fever infection (De La Chapelle, 1929). Considerable pathologic involvement is associated with the disease by evidence too scant for conviction.

The solution must come as postmortem data accumulate in more considerable volume. Quantities of such matter must be filed away in the

records of pathologic laboratories, but in isolated notations of themselves too doubtfully significant for publication. I recall a case in the John Sealy Hospital at Galveston with clinical and serologic aspects warranting report (Stone). Subsequently it disclosed complications with pathologic changes that support and possibly add something to the general picture I shall present, but of itself this feature established too little for an additional report. Findings of bronchopneumonia, pleurisy, arthritis, a rheumatic type of cardiac disease, congestive cardiac failure and acute cystitis were presented at various times. Paralysis agitans and other central nervous derangement, including a pronounced psychosis, also developed. Autopsy revealed gross and microscopic evidence of atheroma of the aorta, hypostatic pneumonia, acute splenitis, purulent cystitis and pyelitis and atrophy and fatty degeneration of the liver. Permission for examination of the brain was not granted. Such conditions are often associated with undulant fever but also with other diseases, and much of their significance lies in the relative frequency.

Without adequate data to pass on the import of pathologic observations, we must be content for the present with a survey of whatever morbid changes are found described. The resulting concept will not be quite true to the disease, perhaps far from that. One can only hope that it may serve pathologists as a basis for comparison in autopsies and encourage the ultimate evolution of a truer picture.

The most essential item in the general pathologic alteration is a proliferation of cells belonging to the reticulo-endothelial system. This has been elaborated on repeatedly of recent years, and earlier post-mortem descriptions of different phraseology offer nothing inconsistent with it. In many cases the proliferation is described as not only pronounced but as sufficiently distinctive to help differentiate the disease from other conditions.

*Nodular Reactions.*—A more or less definite type of infectious granuloma is suggested, the most striking feature of which is a nodular lesion resembling the tubercle. From many quarters something of this sort is described, though not with the same regularity at autopsy as in infected animals. The respective descriptions of histologic structure lack uniformity, however, and leave doubt as to whether they all define the same pathologic reaction; that is, while similar nodules are mentioned by a number of authors, these lesions are not obviously of the same type.

In some instances the histologic picture resembles that of the tubercle. Confusion with concomitant tuberculosis has arisen, but in most cases this mixture of infection seems to be clearly ruled out. Wohlwill described this lesion most distinctly in a case he studied. He said that the nodules were somewhat smaller than millet seeds and of fairly uniform structure. The latter picture was completely overbalanced by

large epithelioid cells having clear, moderately coarse eosinophil, finely granular, or in any case not generally homogeneous, protoplasm and pale nuclei with scant chromatin. Their shape was rarely round, but as a rule oval, hooked, curved or otherwise irregular. Fairly regularly admixed with these were a number of eosinophil or rarely neutrophil granulocytes. In a few of the nodules were occasional giant cells, some resembling Langhans' cells and some having nuclei scattered irregularly. Fat was distributed over the epithelioid cell bodies in fine droplets, making the nodules conspicuous in sudan-stained preparations, but whether this had any importance Wohlwill could not yet say. There was no tendency to caseation.

Points of similarity occur in the histologic structure as described by others. The cellular aggregation is not always in defined nodules (Rössle), nor do defined nodules always consist of a similar cellular aggregation. Some lesions with a superficial resemblance to the tubercle are quite dissimilar in microscopic structure. Gregersen and Lund described nodules consisting of simple fibroblasts and lymphocytes scattered in the fibrous layer, without epithelioid cells. Wohlwill cited Löffler and von Albertini as having found in the pulp of a spleen some older and partly cicatrized nodules that contained plasma cells and were surrounded by fibrinous exudate. Rothenberg described a case of perisplenitis which showed translucent nodules of pinhead size, with hyaline change, scattered over the splenic surface.

While these nodular reactions have been defined most clearly in the spleen, they are also encountered in the lymph glands, bone marrow and other tissues. Hansmann and Schenken described meningeal nodules consisting of hyalinized connective tissue infiltrated with chronic inflammatory cells, large mononuclears surrounded by a collar of lymphocytes. In a patient with undulant fever and tuberculous infection Amoss (1931) found peritonitis simulating tuberculosis. There were white papules softer than tubercles and with a histologic resemblance to them, containing giant cells and lymphoid infiltration. Such a condition has points of similarity to the more marked reaction that sometimes follows a positive allergic cutaneous reaction for undulant fever. The induration, instead of disappearing after a few days, develops into a local granulomatous lesion with a grayish point that suggests underlying pus but on incision produces no pus or bacteria (Giordano).

This tuberculous structure appears much more uniformly and distinctly in infected animals. Gregersen and Lund interpreted findings in man by their approximation to the results of experimentation with animals. The patches they saw in animals showed a structure more like tuberculosis, with a predominance of epithelioid fatty granular cells, while corresponding human lesions showed simple fibroblasts.

In guinea-pigs the resemblance to a tuberculous reaction is so close that it has confused the interpretation of laboratory inoculation tests. Fleischner and Meyer (1917) suggested that earlier testing of milk supplies may have erred because the anatomic lesions in guinea-pigs infected with bovine abortion disease were mistaken for those of tuberculosis. Schroeder and Cotton found that lesions mistakable for tuberculosis are caused by some other organism present in milk, but one not readily identified from their description.

Histologic observations in guinea-pigs are well described by Theobald Smith (1926). In the spleen and lymph nodes a proliferation of endothelial type cells develops, resembling the cells focalized by tuberculosis but not undergoing the same retrogressive changes. They retain stains feebly and give the impression of rarefied areas. This proliferation was not excessive with strains originating from bovine abortion, to which the animal was only moderately susceptible. Polymorphonuclear leukocytes appeared rarely at the center of the focus, when of maximum size, and necrosis practically never. With human strains of greater virulence for guinea-pigs, the foci began similarly, but in two or three weeks they enlarged until they were grossly visible as yellow masses, 2 or 3 mm. in diameter, sharply outlined but not raised. Infiltration with polymorphonuclear leukocytes paralleled the enlargement of the endothelial focus, and a central necrosis sometimes developed. The entire gland was eventually converted into an enlarged firm mass, with coalesced foci. The inconstancy with which a like manifestation has been encountered in man may have some analogy with this contrasting degree of reaction of guinea-pigs to different strains. The tissues may be regarded as insusceptible to the tuberculous reaction in most subjects but are driven to it in the occasional least refractory instances, their reaction then comparing with the milder ones in guinea-pigs. This is only a conjecture, visualizing significance in observations as yet extraordinary. Experiments with inoculation of monkeys have led to the development of similar nodules. One 2 mm. nodule in the lung was described microscopically as a focus of small round cells, and a 6 mm. nodule as one of reticulo-endothelial cells and leukocytes, suppurating at the center (Huddleson and Hallman, 1929).

Necrosis is characteristically absent in the nodular areas in man and occurs more in other organs than in those showing this type of reaction. Conspicuous necrosis of hepatic cells marked the involved patches of that organ in Wohlwill's case, leaving little but an empty framework of connective tissue and persisting capillaries. In contrast with the splenic condition, epithelioid cells were seldom found here. Bassett-Smith (1922) noted a necrosis of the hepatic tissue yielding a positive culture without true pus.

The suppuration described in pronounced nodular lesions in guinea-pigs is likewise absent in man, but suppuration is by no means rare as a complication of undulant fever even in apparently pure infections. In one patient a subdiaphragmatic and hepatic abscess developed (Eyre and Fawcett). A large cavity extended into the liver with necrosis of the organ's substance, and an aspirated specimen yielded the bacillus of undulant fever in pure culture. Abscesses are also reported at other sites, for instance, in the anterior mediastinum (Hardy, Jordan, Borts and Hardy) and in the iliac fossa (Tilghman).

The polymorphonuclear defense is evidently not prominent. The blood picture almost uniformly reveals a decrease of polymorphonuclear and an increase of mononuclear cells. The two changes may result in the raising or lowering or in essentially no change of the total white cell count. Simpson and Fraizer found a slight leukopenia in three fourths of their cases and a normal count in the others. Amoss (1931) found that leukocytosis was the usual response, with 30, 40 or even 80 per cent lymphocytes. Diminution in the number of polymorphonuclears seemingly results from their destruction rather than from bone marrow inhibition, since the proportion of immature nonfilament type cells is increased (Gallagher).

An actual lymphocytosis occurs quite generally and irrespective of the type of organism producing the undulant fever. In the cases of caprine origin early described in Texas, Gentry and Ferenbaugh reported a differential leukocyte count of 45 per cent polymorphonuclears, 47.6 per cent small mononuclears, 4.4 per cent large mononuclears, 2.2 per cent transitionals and 0.8 per cent eosinophils. Similar mononucleosis appears in tabulations of the epidemic in Phoenix that developed subsequently (Watkins and Lake). When Keefer first attributed undulant fever to the abortus type of organism, he found on repeated counts that the lymphocytes rose to as high as 52 per cent and the large mononuclear-transitional group to 22 per cent. Of course there are exceptions. In one diagnosed case there was a leukocytosis of 16,000, of which the polymorphonuclears constituted 71 per cent (Broadbent).

*Spleen.*—The splenic tissue exhibits greater pathologic changes than any other. It was in parts of the spleen that the tubercle-like formations were presented most distinctly. Wohlwill (1932) found them to be most clear and numerous in the malpighian corpuscles, and not in the splenic pulp. Gregersen and Lund described nodules in the pulp, and so did Löffler and von Albertini as cited by Wohlwill. Other microscopic changes have little evident significance. The occurrence of accumulations of certain cells is in accord with observations elsewhere. In keeping with the blood formula, a preponderance of large lymphocytes and large mononuclears are described in splenic smears and in section.

their infiltration about the vessels (Archibald). Macrophages are reported in the sinuses, in which red blood cells and pigment and multi-nucleate giant cells are engulfed (Rothenberg).

The most obvious and general gross change in the spleen is an enlargement. The early cases described by Hughes (1897) were so characterized, and many others since have shown that condition. Muir's textbook of pathology hardly mentioned undulant fever except in connection with a splenic tumor. This author gave the average weight of the organ as a little over 1 pound (0.5 Kg.), with a tendency to progressive increase as the disease progresses. The degree of enlargement varies extremely. The weight is frequently normal, and sometimes, on the other hand, it reaches 1.58 Kg. (Bassett-Smith, 1922). Schottmüller described a patient with an extraordinary swelling, the anterior pole extending to the navel. One of the outstanding physical signs of the condition is a palpable spleen. Manson-Bahr and Willoughby and many others have emphasized it in describing their cases. In the tropics this has confused the splenic index of malaria (Hislop). Infection with the abortus as well as with the melitensis type of organism is characterized by this variable degree of enlargement. Hardy (1929b) found the spleen enlarged in 37 of 125 cases with infections from cattle and swine.

The enlargement results from lymphoid hyperplasia. In his early descriptions of the disease, Bruce (1889) noted that the malpighian bodies were enlarged owing to an increase in the number of round lymphoid cells. This has been generally confirmed since (Strong, 1931). One illustrative record shows somewhat prominent trabeculae and a moderate increase in the pulp (Strong and Musgrave), and many others give similar reports.

The earliest descriptions suggest septic softening (Bruce, 1889). Hughes (1897) described the tissue as appearing almost diffluent, even broken-down and rotten or like a large clot of venous blood. To subsequent pathologists this softening has not seemed so characteristic, and its occurrence has been attributed in part to a postmortem change. The organ has often been found quite firm, perhaps fibrous (Archibald, Tyndale and Viko). Bassett-Smith (1922) suggested an increasing tendency to firmness as the disease becomes chronic. The enormous spleen in Schottmüller's case, several months advanced, was quite solid. Löffler recorded that the diagnostic splenic tumor is larger and more solid than the spleen in typhoid fever.

Passive hyperemia of the spleen is common. Intense congestion on section, with sinuses enormously distended with blood, was observed by Bruce (1889) and subsequently by many others. Incidental observations include small hemorrhages (Bassett-Smith, 1922), organized

thrombi in the trabecular veins (Gregersen and Lund) and anemic infarctions (De La Chapelle). In Rothenberg's case the surface of the spleen showed an acute localized process taking the form of tubercle-like nodules. Perisplenitis is more often encountered in its later effects. In one case tough sheetlike and stringlike adhesions bound the spleen to the diaphragm and to the peritoneum externally (De La Chapelle).

*Lymph Nodes.*—Changes analogous to those in the spleen occur in the lymph nodes. There is likely to be hyperemia of the capsule (Eyre, 1908) and of the gland on cut section (Strong and Musgrave). Enlargement is not infrequent. Beginning with Bruce (1889) many authors have mentioned a swelling of certain lymph nodes, usually including the mesenteric. Occasionally this enlargement is general (Wainwright, 1929b). Palpable and often painful nodes, sometimes located in the neck, may occur early or late in the disease and have sometimes been regarded as diagnostically significant (Rodriguez). The enlargement results from reticulo-endothelial hyperplasia. Descriptions have disclosed this most distinctly along the course of the sinuses (Hardy, Jordan, Borts and Hardy); irregular compression of the sinuses has been known to result (Rössle).

The nodules reminiscent of tubercles have been found well defined in the lymph nodes. Wohlwill encountered them most numerously in the periaortic nodes, their character being like that of the lesions he described in the spleen. Rössle described a distribution of large cells through the lymphoid tissue, in an ill-defined aggregation near the margin but gathered into nodular accumulations deeper in the gland. He noted the resemblance to early stages of tuberculous infection. De La Chapelle found occasional minute white points suggesting miliary abscesses in swollen and edematous lymph nodes. Histologic changes of this character might in some cases contribute to the laboratory diagnosis. In Cruickshank and Cruickshank's case, therefore, a gland was excised, but only lymphatic hyperplasia was found.

As in the spleen, necrosis is exceptional. Katsch described to Wohlwill a reactionless, purulent infused necrosis in numerous groups of lymph nodes. Eyre (1908) mentioned the occasional presence of semi-fluid purulent content in mesenteric nodes. Necrosis and ulceration have occurred similarly in the follicles of the intestine. Bruce (1888) mentioned a few such instances in Peyer's patches, but he noted them only as exceptions to qualify the general observation that ulceration does not occur. The rarity of ulceration in Peyer's patches has always been recognized in the differentiation of undulant from typhoid fever. As a rule, follicles, like nodes, have shown no other change than a slight swelling and cellular hyperplasia (Strong and Musgrave),

Hughes (1897) found patches of intestinal hyperemia in nearly all the cases of his series. As a rule, the alimentary tract shows nothing except this. Ulceration occurs only rarely in the mucosa and then, as a rule, elsewhere than about the lymph follicles. Mention has been made of aphthous patches in the mouth (Müller), of peptic ulcer (Amoss, 1931) and of undermined ulcer in the ileum (Bruce, 1888). Intestinal irritability on roentgen examination has suggested ulceration of the large intestine (Griffin). This, like infection of the gallbladder which also occurs (Amoss, 1931), may help to explain the abdominal pain and tenderness occasionally prominent in the symptoms of undulant fever. Auerbach (1932) mentions a case with symptoms at times of gastritis or gastric ulcer and again of appendicitis or cholecystitis.

*Liver.*—The liver more than any other organ shows a tendency to necrosis. A degeneration about the central veins (De La Chapelle) with centrolobular necrosis (Hardy, Jordan, Borts and Hardy) has been described. Another case showed this process advancing farther until in patches only the empty framework of tissue remained (Wohlwill). In one case a large hepatic abscess was present (Eyre and Fawcett).

Hepatic tissue fails to show at all distinctly the cellular aggregations that sometimes characterize the spleen and a few other organs. In Wohlwill's case they were merely indicated in the affected patches of hepatic tissue. Epithelioid cells were seldom found and then had plenty of room, in contrast with the compressed aggregates elsewhere. Infiltrations of small round cells are frequently mentioned; often they are situated in the interlobular tissues (Bruce, 1889). Increase in connective tissue also occurs in the interlobular areas (Strong and Musgrave).

In many cases the liver is enlarged, though not as markedly as the spleen. In one case the organ was enlarged to 2.94 Kg. (Bassett-Smith, 1922). The enlarged, firm, nutmeg-like liver depicted in a random description (Tyndale and Viko) seems fairly typical. Passive hyperemia develops as in other organs. Hemorrhages occur; in one case they were minute and superficial just beneath the capsule (Strong and Musgrave). Cloudy swelling and fatty change are frequently noted.

*Kidneys.*—Changes in the kidney are pronounced only with complicating nephritis. This complication has been on record since Bruce's work (1889), and with renal insufficiency and uremia has sometimes been the cause of death (Baastrup, 1928). Hemorrhagic cases are described. In that of Strong and Musgrave there were hemorrhages throughout the viscera, and the kidneys were dark and extensively hemorrhagic. In another there were scattered extravasations through the cortex and about some of the glomeruli, with a beginning atrophy of the latter and infrequent hyalinization (De La Chapelle). In the latter case some yellowish specks suggestive of miliary abscesses and

an infiltration of small round cells about some of the Bowman's capsules were also mentioned. Except in instances of nephritis the kidneys show no significant changes. Not infrequently there is some enlargement, hyperemia or cloudy swelling.

*Genital Tract.*— Any involvement of the genital tract naturally attracts attention, since uterine inflammation is the chief manifestation of the infection in cattle and swine. When the organism of contagious abortion was found implicated also in human disease widespread speculation arose as to whether it might be responsible for spontaneous abortion in women. Isolated cases have suggested this. A physician wrote of a patient with a positive agglutination test who aborted twice, the placenta showing a calcified area the first time and necrotic areas full of decomposed blood clot the second time (Harris). Again, an organism culturally like the abortus bacillus appeared in the discharges of an abortion, though this did not occur in a subsequent series of 50 similar cases (Whitehouse, 1929). In another series of 48 cases in which the Wassermann reaction was negative but which were otherwise unselected, cultural examination produced the organism from only a single case, one not studied for signs of undulant fever, though specimens of the fetus or placenta or both were cultivated in all (Carpenter and Boak, 1931). The serums of 23 aborting women similarly proved negative to agglutination test with the abortus organism as antigen, though in 1 case the aborted blood gave a positive reaction (Cornell and De Young). In Gray's series of 62 women whose serums agglutinated the organism, on the other hand, 15 had had one or more abortions. The question of a relationship between spontaneous abortion and undulant fever remains open.

Pathologic changes such as those leading to abortion in cattle are not described in human cases. In the cow this inflammation is quite pronounced and characteristic. A yellowish or brown exudate forms between the uterine mucosa and the chorion; it varies in character from mucopurulent to tenacious and gluey and is composed of detritus, leukocytes and degenerating epithelial cells. The uterine mucous membrane is frequently swollen, hyperemic or hemorrhagic and is roughened by serofibrinous exudate or even shows necrotic areas (Mohler and Traum). The abortus organism invades and densely fills the epithelial cells of the chorion, and these show enlargement and vacuolation of the cytoplasm (Smith, 1919).

The study showing closest approximation to this in women antedates the discovery of undulant fever of the abortus type. It was concerned with possibilities of human contagious abortion. Three women had aborted, the fetus being expelled and the placenta removed by curet. A discharge, brownish with blood and thick, continued but

the abortus organism was not recovered (De Forest). Nothing of the sort is described in patients with undulant fever, though there has been a suggestion of the infection localizing at this site. One patient with a frank case of caprine origin aborted in the fifth month of gestation, and serum of the dead fetus showed a positive agglutination reaction (Samut). Congenital undulant fever was noted in the days of the Mediterranean Fever Commission (Williams); symptoms developed in a baby after delivery and without contact with the sick mother.

The principal morbid changes described in the female generative system involve the ovary. Oophoritis has been associated with peritoneal inflammation (Amoss and Poston). Cysts have been described: In one case they were small and hemorrhagic and located in the left ovary (Amoss, 1931) and in another they persisted so chronically as to yield a positive culture after six years (Wainwright, 1929a). A number of patients with undulant fever are said to have become sterile (Giordano and Ableson).

Any of the male generative organs, most often the testes, may show involvement. According to one estimate (Wainwright, 1929b) a mild orchitis occurs in 20 per cent of the cases of melitensis infections and in 4 per cent of those of abortus type. Testicular changes have been listed among the minor physical signs to be considered in diagnosis (Darbois, 1910). One pathologic description noted the evidence of acute interstitial orchitis and scattered areas of fibroid atrophy with vacuolar degeneration of the epithelium (Rothenberg). The epididymes may also be inflamed (Müller). Cases have been described in which prostatitis and vesiculitis, sometimes suppurative, have undoubtedly resulted from undulant fever (Simpson and Fraizer, Herbert).

*Chest.*—Changes in the chest are even less distinctive than those in the abdomen and usually represent complications. The pleural membrane is inflamed more often than the pericardial or peritoneal membrane. Fluid in the body cavities has been repeatedly mentioned since the earliest autopsies, usually the clear fluid of a transudate (Eyre, 1908) but often an exudate of inflammatory origin. Subsequent adhesions are not at all uncommon. Hughes (1897) encountered pleural adhesions in 6 of his cases of acute undulant fever, old adhesions in 3 and recent ones in 3. Fairly extensive obliteration of the pleural cavities is often recorded (De La Chapelle). Indication of this involvement by a homogeneous dulness in the roentgenogram (Jenkins, 1929) may find a prominent place in the clinical picture presented.

*Lungs.*—Bronchopneumonia readily complicates the disease, and in many cases its manifestations have been among the outstanding features. Any number of postmortem examinations have shown a greater or less degree of lobular consolidation. This occurrence, like the pleural

involvement, brings in the diagnostic roentgenogram. The film may show a general increased density and some mottling or peribronchial infiltration (Carpenter and Merriam). In certain instances the clinical manifestations have simulated tuberculosis (Bethoux). In one outbreak 15 of 75 cases of undulant fever showed pulmonary localization, and several were first recorded as tuberculosis (Vanni).

Mention is frequently made of hyperemia about the bases of the lungs; pulmonary edema is also noted. A pulmonary abscess has been described, though the authors (Hardy, Jordan, Borts and Hardy) could not determine whether it was due to the primary infection or to some secondary invasion favored by the lowering of resistance.

*Heart.*—The most striking of the cardiac findings is a vegetative endocarditis. This complication is encountered again and again. Hughes (1897) described vegetations on the mitral valve in 3 cases, but he thought that in one case they were probably and in the others presumably of previous origin. The more recent vegetations showed infiltration with round cells and the older ones an organization into fibrous tissue. Scott and Saphir (1928) also thought that the endocarditis they found in a case of septicemic undulant fever was probably of other causation, very likely rheumatic. The chordae tendineae of the mitral valve were thickened, shortened and adherent, and the leaflets thick, firm and covered with small, yellowish-gray, friable vegetations. The leaflets of the aortic valve were also retracted and rigid; they showed friable vegetations on their free margins and an adherent, soft, round, reddish-gray thrombus.

De La Chapelle described a vegetative and ulcerative endocarditis as being very likely the main seat of infection in a case of undulant fever and as undoubtedly caused by the organism. The two anterior cusps of the aortic valve were almost completely destroyed and replaced by a cream-colored, granular, fused mass of soft and friable vegetations partially blocking the orifice. During the four months of the disease, certain symptoms like those of a subacute streptococcic endocarditis had been noted. Others also have been inclined to think that the valvular inflammation they describe is the result of an attack of undulant fever (Heiberg; Hardy, Jordan, Borts and Hardy).

The myocardium rarely shows any significant abnormalities. Pallor is sometimes recorded and in chronic cases, fatty degeneration. In De La Chapelle's case of endocarditis it presented, besides a moderate cloudy swelling and granular degeneration of muscle fibers, a slight infiltration with large and small round cells. The blood vessels about the various sites of infective localization through the body have shown analogous cellular infiltrations. These are usually of small round cells, forming well defined aggregations in the venous wall (Wohlwill).

Thrombophlebitis has been on record so long (Cantani, 1914) and so prominently that it is surprising that deaths from emboli are not reported (Wohlwill).

*Hemorrhage.*—The tendency to hemorrhage is pronounced; at times it is generalized. Extravasations have been mentioned in connection with organs already described. Petechiae occur in the serous and mucous membranes and in the skin (Scott and Saphir, De La Chapelle). Bleeding is one of the clinical signs of the disease. Angle described a persistent epistaxis, ulorrhagia and hematuria, as well as a distribution of petechiae over the surface of the body. He found the bleeding time markedly prolonged, a platelet estimate of 16,000, and the clotting time three minutes.

*Skin.*—Changes in the skin have been frequently recorded clinically but not at autopsy. They are largely blood vascular. A roseola assumes importance because of clinical similarity between this disease and typhoid fever. Red, macular, scaling lesions, more or less distributed over the body, appear in about 5 per cent of the cases, according to Simpson and Fraizer. In some cases they present a striking resemblance to rose spots. In one case, eventually fatal, there were successive crops until by the seventh day the body was nearly covered (Duffie). An erythema multiforme has been encountered repeatedly, and once an erythema exudativum was seen (Müller). Allergic manifestations also are noted in the skin. Exposure of the hands to vaginal discharges of cows sick with contagious abortion has commonly led to an itching rash among veterinarians. This rash may consist either of irregular blotches made up of minute reddish points or of discrete red papules which in a few days change to brown (Huddleson and Johnson, 1930).

*Rheumatic Symptoms.*—Rheumatic symptoms enter into the disease too often to be disregarded. Some of the patients first found to have undulant fever in this country had been sent to the Army and Navy Hospital at Hot Springs for articular rheumatism (Curry). Cases have since presented symptoms in the joints with extreme frequency; a third of those in the Dayton series showed those symptoms (Simpson, 1930). In a few such cases morbid changes of the joint are definitely evident. Swelling and other signs of acute arthritis localize about a particular joint, the elbow for instance (Wellman, Eustis and Schuchet). Baker isolated the organism from fluid aspirated from the knee after months of intermittent effusion into that joint. More often the condition in the joint is defined symptomatically only.

*Nervous System.*—Much the same might be said of the frequent neuralgic and other symptoms referable to the nervous system. In one series, with arthritis dominant, all the cases showed some such nervous symptoms: neuritis of shifting location, tremor, mental depression, exces-

sive tension or insomnia (Sensenich and Giordano). Much of this has been attributed rather indefinitely to a toxic disorder of the nervous system incidental to the general sepsis. Old cases with temporary paralyses point to some such pronounced disturbance. R. de Nunno was cited (Bassett-Smith, 1922) as stating that dead as well as living cultures of the organism produce degenerative changes of the nerve cells, with breaking up of the fibrils and leukocytic infiltration. This is less in the cord than in the cerebrum and medulla; it is marked also in the peripheral nerves. Herpes zoster has complicated the disease with implication of the posterior root ganglions of the fourth or fifth lumbar nerves (Bassett-Smith, 1920).

*Bone.*—Pronounced evidences of nervous disorder have been attributed in some instances to involvement of the vertebrae. Rawak and Braun cited several cases in which destructive changes of lumbar vertebrae, shown in roentgenograms, led to radiating pains in the back. In a case of their own, such pains and also atrophy of the muscles of the shoulder resulted from vertebral destruction, principally in the cervical region. Destructive changes of the vertebrae, sometimes with suppuration, are mentioned repeatedly. Trotta described suppurative changes simulating those of Pott's disease. According to Kulowski and Vinke others find both in man and in cattle an association of pronounced formation of abscesses with destructive change of the lumbar vertebrae. Abscess matter from their own case yielded a culture of the bovine organism.

Destruction of bones in undulant fever is by no means limited to the vertebral column. Weil encountered an arthritis following the disease and with it a metatarsal osteitis that caused severe swelling, pain and discoloration of the foot. Wohlwill cited Smith and Fabyan to the effect that the bone is destroyed from the marrow outward, and he therefore thought that the changes he himself saw in the marrow perhaps constituted the original osteal lesion. The pathologic structure he described consisted of small nodules like those in the spleen and lymph nodes but shaped into proliferal branches that extended along the dividing walls between droplets of fat.

*Meningeal Involvement.*—Occasionally the disease has been complicated by meningitis. Hansmann and Schenken carefully described one such instance in a case of undulant fever of porcine origin. The lepto-meninges about the anterior and central portion of the cerebral hemispheres showed the greatest involvement, mainly along the vessels. In this region there appeared many grayish-white, tubercle-like structures. Much of the inflammatory change was obscured by hemorrhage, as a ruptured aneurysm of the basilar artery had filled the subarachnoid space at the base of the brain with blood. One of the tubercle-like

structures showed irregular masses of hyalinized connective tissue infiltrated with chronic inflammatory cells, large mononuclears surrounded by a collar of lymphocytes. Another was necrotic and contained polymorphonuclear leukocytes. The nodules were apparently proceeding from necrosis to connective tissue hyalinization. Further microscopic changes included a thickening of the pia and arachnoid membranes, with cellular infiltration and proliferation of fibrous tissue. Abundant lymphocytes, plasma cells and large mononuclears were in evidence, some of the lymphocytes appearing in perivascular accumulations. During life, the outstanding observation on the spinal fluid had been mononuclear pleocytosis, the counts recording 300 cells with 36 per cent lymphocytes and 271 cells with only 12 per cent polymorphonuclears.

Hansmann and Schenken found records of 3 proved cases of meningeal complication in the melitensis type of infection, but none prior to their own in the abortus type. Bingel and Jacobsthal subsequently reported a case in which they recovered the abortus organism from the spinal fluid. Some involvement of the brain and meninges appears to be less infrequent than one would presume from observations on encephalomeningeal reactions (Roger). Brachiofacial dysesthesia and other sensory or even motor disturbances follow a consequent irritation of the middle cerebral artery with spasms, the findings being characteristic of undulant fever. In Hughes' (1897) early series of cases of undulant fever, the meninges had been described as usually hyperemic, and in some instances there was effusion into the ventricular spaces.

#### IMMUNE REACTIONS

The literature does not tell much about the body's natural resistance to this infection. Susceptibility to attack by the respective types of the organism differs markedly in degree, as pointed out in connection with bacteriologic observations. The lesser liability to infection during childhood is due in part to the low pathogenicity of the one type likely to reach children through milk. This protection has been attributed in part also to a lowering of susceptibility by biologic immaturity (Dietrich and Bonyngé). Calves, fully exposed to the variety of the organism infective for them, exhibit the same relative immunity (Simpson, 1930).

From the first clinical observation has suggested an acquisition of at least relative immunity following an attack (Hughes, 1897). Conference of immunity by inoculation has been repeatedly and extensively tried, both prophylactically and therapeutically. The experimental use of antiserum to induce passive immunity has failed. Wright tried goat serum ineffectively and then inoculated horses, but the serum from immunized horses proved equally discouraging in various hands (Fitzgerald and Ewart; Hitchens). Current work with the serum of a goat immunized by heavy doses of vaccine detoxified with nitrous acid seems

a little more encouraging in its preliminary report (O'Neil). The dim prospect of effective serum therapy is not brightened by observations on the effect of the serums on immunized rabbits and guinea-pigs. While high titers of these serums dubiously protect guinea-pigs from experimental infection they do not at all modify the termination of an established infection (Gwatkin, 1933).

The possibility of building an active immunity by means of vaccines has been considered since the days of the Mediterranean Fever Commission (Eyre, McNaught, Kennedy and Zammit). Castellani seemed to find a prophylactic effect and incorporated selected strains of the organism of undulant fever in typhoid-paratyphoid vaccines (Castellani and Taylor). Various attempts have been made to improve the antigenic quality through the technic of preparation, as killing by chemicals instead of by heat (Gwatkin). Many have been encouraged by the results of their inoculations, vaccinated comparing favorably with nonvaccinated groups in some cases (Dubois and Sollier). Proofs of efficiency have not been widely accepted, however, for any prophylactic vaccine. This line of experimentation has brought similarly unsuccessful results with contagious abortion among cattle (Cotton, 1924).

The inadequacy of all known forms of treatment has led to extensive experimentation with therapeutic vaccines. When treatment with stock vaccine proved discouraging, the organisms obtained in blood culture were employed for inoculation (Kennedy, 1910; Owen and Newham, 1915). High hopes were aroused by early reports, but as these failed to materialize, the treatment has lost ground. Investigators still hope to produce an effective vaccine antigen. Concentration by special method and intramuscular administration have been tried (Schilling, Magee and Leitch) and the broth culture filtrate brucellin has been used, administration being gaged by phagocytic activity of the patient's cells (Huddleston and Johnson, 1933). The specific effectiveness of any immunization procedure remains to be demonstrated.

The curative effects early described are relegated more and more distinctly by subsequent work to the nonspecific. In one series of cases good results were found to follow only an intense general reaction (Cambessédès and Garnier). This led the authors to recommend large doses, and the question of a protein reaction arose. In recent years several clinicians have employed a typhoid-paratyphoid vaccine for protein shock therapy. This reduces the fever in about the same manner as the specific organism vaccines (Budtz-Olsen; Miller; Manson-Bahr, 1933).

Serologic changes of greater moment in undulant fever are the evidences of immunization that denote the infection. These have diagnostic significance far exceeding that of other clinical and laboratory findings. They were first thought to carry a practical prognostic indica-

tion as well, persistently low or declining agglutination titer of a patient's serum suggesting a bad prognosis (Birt and Lamb; Bassett-Smith, 1902).

The disease was early differentiated from others by agglutination (Wright and Smith, 1897), and this test has remained the principal diagnostic procedure. For considerable significance the reaction must occur with the serum highly diluted. Lesser agglutination potency is acquired by the serums of many persons independently of clinical undulant fever. This was mentioned in connection with the occupational distribution of the disease, veterinarians commonly having agglutinin in their serum even to a 1:100 titer or higher (Huddleson and Johnson, 1930). This mild agglutination property has been attributed to a sub-clinical infection or even to the entry of organisms without infection. Continuous ingestion by monkeys of abortus organisms in small numbers results in a mild and unrecognizable yet immunizing infection (Meyer and Eddie). In one series of 500 persons with various diseases, 58 of the serums agglutinated the organism in dilutions ranging from 1:5 to 1:40 and 1 agglutinated it at 1:320 (Evans, 1925). Other series show varying degrees of agglutination similarly in 5 per cent or more (Hull and Black).

Low dilution with positive reactions may eventually assume diagnostic dependability provided a high ratio of the agglutinins is soluble in carbonic acid, according to Gray's experiments on rabbits. In animals recently immunized the specific agglutinin occurs mainly in the soluble fraction of the serum, but this portion decreases much more rapidly than that in the insoluble fraction. Scant agglutinin might therefore be attributed to an active infection if it occurs in the soluble fraction or to a previous immunizing incident if in the insoluble fraction. It seems unfortunate to disregard the low dilution reactions as largely as one now must, for a high or even a low titer of agglutination does not occur uniformly with known serum of undulant fever. A proportion of positive cases diagnosed by blood culture and serologic test fail to give the agglutination reaction just as a proportion fail to yield a blood culture of the organism (Carpenter, 1926).

Type specificity of the agglutination reaction is not at all distinct. The infecting type of the organism is likely to react to a more or less higher titer than other types. In one series of cases of caprine origin, 15 serums agglutinated the melitensis organism at 1:1,000 or 1:2,500 dilution and the remaining 4 at 1:500, while they agglutinated the abortus organism either not at all or not above 500 (Phease). De Korte's (1924) case of abortus fever gave no agglutination with the melitensis but a positive reaction with the abortus type. On the other hand, the serum in a case of undulant fever in the Rio Grande valley, contracted after drinking raw goat's milk regularly, reacted to the highest titer with

the abortus variety of the organism (Stone); a culture for typing could not be obtained. To a great extent patients' serums agglutinate indiscriminately any of the three types of organism.

Undulant fever serum is likely also to agglutinate the tularemia organism (Francis and Evans). In man and also in inoculated animals there is cross-agglutination between these two organisms, though the titer is ordinarily higher for the specific one. Occasionally patients' serums, agglutinating both at nearly equal titer, have required absorption of agglutinin for diagnosis (Carpenter, 1926). This antigenic relationship has influenced one classifier of bacteria to include the tularemia organism tentatively in the Brucella rather than the Pasteurella group (Topley and Wilson, *b*).

Immune reactions other than agglutination are readily demonstrable, though without conspicuous advantage in diagnosis. The precipitin reaction is applicable, but has nothing to recommend it over agglutination or complement fixation, except perhaps technical adaptability to the individual laboratory (Schlesmann). Complement fixation was first employed to detect infection in animals and proved as applicable to human infections, giving results generally parallel with agglutination (Larsen and Sedgwick). It becomes positive somewhat more slowly in experimentally infected goats than does the agglutination test, ten days elapsing before complete fixation takes place as against five for agglutination at 1:40 and six at 1:500 (Mohler and Eichhorn). The procedure does not overcome the outstanding errors of the agglutination test. It likewise frequently gives positive reactions in healthy persons and, according to Sedgwick and Larsen, in infants soon after they are weaned from the breast. In no respect has it proved preferable, but it is valuable for confirmation of a diagnosis when the agglutination reaction and the clinical findings conflict (Morales-Otero and Monge).

With regard to opsonic influence on phagocytosis by polymorphonuclear leukocytes, one recalls the lymphogenic character of the antigen and does not expect a great deal. A decrease is noted in the number of phagocytes and in their individual activity, but generally there is a rise in opsonin during convalescence (Bassett-Smith, 1922). In a recent paper an indication of progress toward recovery is ascribed to the phagocytic activity demonstrable in citrated samples of blood. Phagocytosis of the organism by polymorphonuclear leukocytes is thought expressive of immunity, and the lack of such activity in conjunction with a negative cutaneous reaction, of susceptibility (Huddleson, Johnson and Hamann). The reaction has been employed practically for gaging the progress of immunization (Sander).

Cutaneous hypersensitivity to the infective agent, analogous to that in tuberculosis, was observed in guinea-pigs by Fleischner and Meyer (1918). They applied the test also to man for ruling out infection in

a series of infants. An allergic test with intradermal inoculation soon found diagnostic use (Burnet, 1922). Various antigenic preparations were used. Trenti (1923) inoculated the filtrate of a twenty day broth culture. Preference settled on a suspension of organisms from solid mediums. Giordano suspended the growth in saline solution to a density of 1:1,000 by silica standard, killed it by heat, injected 0.2 cc. intradermally, and looked for a local inflammatory reaction after from twelve to forty-eight hours. He obtained a pronounced positive reaction in 25 cases of undulant fever and a negative in all but 1 of 100 healthy controls. Early results showed an approximate parallel with those of agglutination (Trenti, 1925). Cross-tests between bovine and caprine strains were as strongly positive as the direct test in infected guinea-pigs (Fleischner, Meyer and Shaw) and in man (Bua).

One significant divergence in result from the serologic test appeared, in that the reaction remained positive much longer after subsidence of the active infection. Of 365 tested persons in one series, 27 who gave reason for the belief that they had or had had undulant fever gave a distinctly positive reaction to the cutaneous, but, in some instances, a negative one to the agglutination, test (Levin). Because the positive reaction fails to distinguish between present and past infection some authors attach less importance to its positive evidence of infection than to the negative evidence ruling infection out (Yeckel and Chapman). Serologic tests are preferred for the diagnosis of undulant fever, but this one confirms the doubtfully negative findings. Mallory suggests that physicians avoid the cutaneous test in presumable cases of undulant fever because it sometimes leads to a violent reaction, even to extensive necrosis (Cabot).

#### LABORATORY DIAGNOSIS

In view of the outstanding diagnostic value of laboratory studies on undulant fever, this review may appropriately conclude with a résumé of findings most significant in the laboratory diagnosis. First among these is the recovery and identification of the organism or its recognition by pathogenic effect on animals. Immune manifestations are nearly as significant and much more readily observable. Clinical microscopic examination adds to the initial presumption of undulant fever by means of the blood cell formula.

Of all the laboratory evidence, the observation most certainly indicative of undulant fever is recovery of the organism in culture or the infection of animals. Some permit only a tentative diagnosis on serologic test and clinical findings (Awe and Palmer). A culture is obtained most readily from the blood. The urine frequently contains organisms, though it is likely to prove positive only after repeated cultivation of samples, even with improved methods now current; the stools or some focal abscesses in exceptional instances yield the organism (Wilson,

1930). Isolation from stool demands a special technic (Amoss and Poston, 1929).

For recovery of the organism a technical procedure commonly approximated of recent years is described by Huddleson, Halsey and Torrey. They make use of liver infusion medium, control of atmosphere, differential inhibition of dye and other devices. Isolation and early cultivation too often present difficulty for the neglect of any favorable influence. Orpen suggested a method of concentration, plating the bacterial and red blood cell sediment of a centrifugated specimen. Colonies may develop within a few days and are then picked for subculture and serologic identification, but a period of two weeks or more of incubation is often required before sufficient growth develops.

The organism was first described as nonpathogenic for laboratory animals, but some years later it was found infective for rabbits and guinea-pigs (Durham). Guinea-pigs proved best suited for diagnostic inoculation; but they rarely die until after from two to three months. The animals may be killed in four or five weeks for autopsy and splenic culture (Carpenter and Boak, 1930). Infection leads to more or less characteristic change in the tissue, already described. The changes are not equally distinct with all the types and strains, and no other evidence of infection than a positive agglutination reaction of the animal's serum may appear (Cruickshank and Barbour). Infected mice yield earlier splenic culture (Hagan), and this method may offer advantages. Rainsford finds the Aleppo hamster more susceptible to infection and better adapted for diagnosis than the guinea-pig.

Among the serologic tests that of outstanding diagnostic value is agglutination. Undulant fever was among the first of the conditions to which this reaction was applied (Wright and Smith). Technical procedures were then described (Wright and Semple) adapting it for general clinical use, which it has had ever since. The serum from cases of undulant fever does not always agglutinate the organism, and that from patients without this infection may agglutinate it. Patients with severe undulant fever have had serums that were negative, or positive only at 1:15 or 1:30 (Carpenter and Boak, 1930). On the other hand, only the serum from undulant fever is at all likely to react in high dilution and it nearly always does react so. The positive agglutination test carries much more conviction if reactive to titers of 100 or above or if confirmed by culture or infection of animals. Diagnosis from less evidence than this is usual in many public health laboratories consulted by Gibbes, but is pointed out to be definitely inadequate.

Altered technical procedure has adapted the agglutination test to special situations. For immediate quantitative result the test is made on the slide, with graduated amounts of undiluted serum reacting against a standardized suspension of heated organisms in 12 per cent saline

solution; reading is from the gross flocculation that takes place (Huddleson and Abell, 1928). Bass' bedside method of agglutination for typhoid fever has likewise been adapted to undulant fever (Lewis). The "abortoscope" Bevan devised for use with cattle is also made use of, a loopful of positive blood clearing a bacterial suspension sufficiently for perception of writing through it (Ross, 1927b).

Since there is considerable liability to accidental infection in laboratories, the use of killed antigen stock instead of living culture assumes practical value when tests are set up by general laboratory technicians. Bevan (1921) devised a method of killing with chloroform, using his killed suspension for agglutination tests and also for vaccine. Other chemical treatments for killing and preservation find wide employment. Formaldehydized, thick milky suspensions are furnished by biologic supply houses; this stock can be diluted as required for the usual gross test. In the hands of bacteriologists, preference is sometimes expressed for the living organism as antigen (Carpenter and Boak, 1930).

The serologic test next in favor is complement fixation. This has the same disadvantages as agglutination, presenting false positive as well as false negative reactions. In one series of 1,000 unselected persons, including only 5 with known undulant fever, the complement-fixation test was positive in 96 and agglutination occurred at low dilution in 78 (Sasano, Caldwell and Medlar). Fixation may well accompany the agglutination test as a confirmatory procedure. The only serologic test to differentiate adequately between types of undulant fever is absorption of agglutinin (Carpenter and Boak, 1930).

None of the changes in the tissues are highly characteristic. Some are reflected in physical signs or in roentgenograms. The only microscopic specimen of significance is the blood smear. Others, such as sections of excised lymph glands, have rarely been considered. Differential diagnosis utilizes the blood formula principally for characteristic mononucleosis. One author suggests that while this method is not diagnostic in itself, it does advance the diagnosis from possibility to probability (Nyfeldt). Possibility of specific utilization of the blood picture is suggested by an experimental hemoclastic reaction; a specific vaccine in inoculated rabbits or in patients with undulant fever reduces the white cell count by over 1,000 cells and leads to inversion of the leukocyte formula (d'Amato, Bossa).

#### BIBLIOGRAPHY

- d'Amato, L.: *Riforma med.* **44**:32, 1928.  
Amoss, H. L.: *Internat. Clin.* **4**:93, 1931.  
— and Poston, M. A.: *J. A. M. A.* **93**:170, 1929.  
Anderson, E. D., and Pohl, J. F.: *Am. J. Dis. Child.* **42**:1103, 1931.  
Angle, F. E. J.: *Kansas M. Soc.* **30**:323, 1929.  
Archibald, R. G.: *J. Trop. Med.* **26**:55, 1923.

- Atwood, G. E., and Hasseltine, H. E.: Pub. Health Rep. **45**:1343, 1930.  
Aubert, P.; Cantaloube, P., and Thibault, E.: Ann. Inst. Pasteur **24**:376, 1910.  
Auerbach, T.: Med. Klin. **28**:1639, 1932.  
Awe, C. D., and Palmer, H. D.: Am. J. M. Sc. **176**:837, 1928.  
Baastrup, V. I.: Ugesk. f. læger **90**:457, 1928.  
Baker, B. M., Jr.: Arch. Int. Med. **44**:128, 1929.  
Bang, B.: J. Comp. Path. & Therap. **10**:125, 1897.  
Bassett-Smith, P. W.: Brit. M. J. **2**:861, 1902; **2**:324, 1904; J. Trop. Med. **17**:93, 1914; **23**:201, 1920; Undulant Fever, in Byam, W., and Archibald, R. G.: The Practice of Medicine in the Tropics. New York, Oxford University Press, 1922, vol. 2, p. 998; Proc. Roy. Soc. Med. (jt. disc., Sects. Comp. Med., Obst. & Gynæc., & Trop. Med. & Parasitol.) **19**:13, 1926.  
Bergey, D. H.: Manual of Determinative Bacteriology, ed. 4, Baltimore, Williams & Wilkins Company, 1933.  
Bethoux, L.: Presse méd. **37**:835, 1929.  
Bevan, L. E. W.: Tr. Roy. Soc. Trop. Med. & Hyg. **15**:215, 1921; Proc. Roy. Soc. Med. (jt. disc., Sects. Comp. Med., Obst. & Gynæc., & Trop. Med. & Parasitol.) **19**:8, 1926.  
Bierring, W. L.: J. A. M. A. **93**:897, 1929.  
Bingel, A., and Jacobsthal, E.: Klin. Wchnschr. **12**:1093, 1933.  
Birt, C., and Lamb, G.: Lancet **2**:701, 1899.  
Blake, F. G., and Oard, H. C.: Yale J. Biol. & Med. **1**:128, 1929.  
Blanchard, R.: Bull. Acad. de méd., Paris **65**:181, 1911.  
Blumer, G.: Ann. Int. Med. **3**:122, 1929.  
Bossi, G.: Riforma med. **45**:211, 1929.  
Boyd, W.: A Textbook of Pathology, Philadelphia, Lea & Febiger, 1932.  
Broadbent, W.: Lancet **1**:76, 1931.  
Bruce, D.: Practitioner **39**:161, 1887; **40**:241, 1888; Brit. M. J. **1**:1101, 1889.  
Bua, F.: Policlinico (sez. prat.) **34**:631, 1927.  
Büdtz-Olsen, J.: Ugesk. f. læger **92**:596, 1930.  
Burnet, E.: Compt. rend. Acad. d. sc. **174**:421 and 973, 1922; **187**:545, 1928.  
Cabot, R. C.: New England J. Med. **208**:1317, 1933.  
Cambessédès, H., and Garnier, G.: Paris méd. **1**:281, 1929.  
Cantani, A.: Policlinico (sez. prat.) **21**:741, 1914.  
Carey, J. D., and Newsom, I. E.: Colorado Med. **26**:320, 1929.  
Carpenter, C. M.: J. Infect. Dis. **39**:220, 1926; J. Am. Vet. M. A. **70**:459, 1927.  
— and Boak, Ruth: Am. J. Pub. Health **18**:743, 1928; J. Lab. & Clin. Med. **15**:437, 1930; J. A. M. A. **96**:1212, 1931; Am. J. M. Sc. **185**:97, 1933.  
— and Merriam, H. E.: J. A. M. A. **87**:1269, 1926.  
Castellani, A., and Taylor, F.: Brit. M. J. **2**:356, 1917.  
Clark, R. H.: New Orleans M. & S. J. **85**:737, 1933.  
Clouston, H. R.: Canad. M. A. J. **28**:535, 1933.  
Cornell, E. L., and De Young, C. R.: Am. J. Obst. & Gynec. **18**:840, 1929.  
Cotton, W. E.: J. Am. Vet. M. A. **62**:179, 1922; Vet. Med. **19**:463, 1924.  
Craig, C. F.: Am. J. M. Sc. **125**:105, 1903; Internat. Clin. **4**:89, 1906.  
Cruickshank, J. N., and Cruickshank, R.: Brit. M. J. **1**:195, 1930.  
Cruickshank, R., and Barbour, W. J.: Lancet **1**:852, 1931.  
Curry, J. J.: J. M. Research **6**:241, 1901.  
Dalrymple-Champneys, W.: Brit. M. J. **2**:604, 1931; Proc. Roy. Soc. Med. **26**:1093, 1933.  
Darbois, P.: Presse méd. **18**:923, 1910.  
De Forest, H. P.: Am. J. Obst. & Gynec. **76**:221, 1917.

- De La Chapelle, C. E.: Am. Heart J. **4**:732, 1929.
- Demaree, E. W.: Kentucky M. J. **31**:343, 1933.
- Dietrich, H., and Bonynge, C. W.: J. Pediat. **1**:46, 1932.
- Dubois, C., and Sollier, N.: Ann. Inst. Pasteur **45**:596, 1930.
- Duffie, D. H.: J. A. M. A. **87**:1830, 1926.
- Duncan, J. T.: Tr. Roy. Soc. Trop. Med. & Hyg. **22**:269, 1928.
- Durham, H. E.: J. Path. & Bact. **5**:377, 1898.
- Evans, A. C.: (a) J. Infect. Dis. **22**:580, 1918; (b) ibid. **23**:354, 1918; (c) U. S. Pub. Health Service, Hygienic Lab. Bull. no. 143, 1925; (d) J. A. M. A. **88**:630, 1927.
- Eyre, J. W. H.: J. Roy. Army M. Corps **8**:113, 1907; Lancet **1**:1747, 1908; **1**:88, 1912; Proc. Roy. Soc. Med. (jt. disc., Sect. Comp. Med., Obst. & Gynæc., & Trop. Med. & Parasitol.) **19**:1, 1926.
- and Fawcett, J.: Guy's Hosp. Rep. **59**:209, 1905.
- McNaught, J. G.; Kennedy, J. C., and Zammit, T.: Great Britain Mediterranean Fever Commission Reports, London, Harrison & Sons, 1907, pt. 6, p. 130.
- Ferenbaugh, T. L.: J. A. M. A. **57**:730, 1911.
- Feusier, M. L., and Meyer, K. F.: J. Infect. Dis. **27**:185, 1920.
- Fitzgerald, E. D., and Ewart, J. H.: Lancet **1**:1924, 1899.
- Fleischner, E. C., and Meyer, K. F.: Am. J. Dis. Child. **14**:157, 1917; **16**:268, 1918.
- Meyer, K. F., and Shaw, E. B.: ibid. **18**:577, 1919.
- Veckii, M.; Shaw, E. B., and Meyer, K. F.: J. Infect. Dis. **29**:663, 1921.
- Francis, E.: Pub. Health Rep. **46**:2416, 1931.
- and Evans, A. C.: Pub. Health Rep. **41**:1273, 1926.
- Gallagher, J. R.: Am. J. M. Sc. **185**:391, 1933.
- Gentry, E. R.: Undulant Fever, in Christian, H. A.: Oxford Medicine, New York, Oxford University Press, 1930, vol. 4, p. 799.
- and Ferenbaugh, T. L.: J. A. M. A. **57**:889 and 1045, 1911.
- Gibbes, J. H.: South. M. J. **24**:126, 1931.
- Gilbert, R., and Coleman, M. B.: J. Infect. Dis. **43**:273, 1928.
- Giordano, A. S.: J. A. M. A. **93**:1957, 1929.
- and Ableson, Marjorie: J. A. M. A. **92**:198, 1929.
- Good, E. S., and Smith, W. V.: J. Bact. **1**:415, 1916.
- Gray, J. D. A.: J. Bact. **25**:415, 1933.
- Great Britain Mediterranean Fever Commission Reports, London, Harrison & Sons, pts. 1, 2 and 3, 1905; pt. 4, 1906; pts. 5, 6 and 7, 1907.
- Gregersen, F., and Lund, T. M.: Hospitalstid. **74**:349, 1931.
- Griffin, W. A.: New England J. Med. **202**:324, 1930.
- Gwatkin, R.: J. Infect. Dis. **48**:381, 1931; **53**:230, 1933.
- Hagan, W. A.: J. Exper. Med. **36**:727, 1922.
- Hansmann, G. H., and Schenken, J. R.: Am. J. Path. **8**:435, 1932.
- Hardy, A. V.: (a) Pub. Health Rep. **43**:2459, 1928; (b) J. A. M. A. **92**:853, 1929; (c) **93**:891, 1929.
- Jordan, C. F.; Borts, I. H., and Hardy, G. C.: Nat. Inst. Health Bull. no. 158, 1931.
- Harris, H. J.: J. A. M. A. **101**:1584, 1933.
- Hasseltine, H. E.: Pub. Health Rep. **44**:1659, 1929; **45**:1660, 1930; **46**:1519, 1931.

- Heiberg, S.: Hospitalstid. **73**:933, 1930.
- Herbert, L. A.: New Orleans M. & S. J. **84**:259, 1931.
- Hill, O. W., and Monger, R. H.: J. A. M. A. **97**:176, 1931.
- Hislop, J. A.: Brit. M. J. **2**:870, 1902.
- Hitchens, A. P.: Am. J. Trop. Dis. **1**:228, 1913.
- Holt, R. F., and Reynolds, F. H. K.: Mil. Surgeon **56**:414, 1925.
- Huddleson, I. F.: J. A. M. A. **86**:943, 1926; J. Bact. **17**:58, 1929; Am. J. Pub. Health **21**:491, 1931.
- and Abell, E.: J. Bact. **13**:13, 1927; J. Infect. Dis. **42**:242, 1928.
- and Hallman, E. T.: J. Infect. Dis. **45**:293, 1929.
- Halsey, D. E., and Torrey, J. P.: J. Infect. Dis. **40**:352, 1927.
- and Johnson, H. W.: J. A. M. A. **94**:1905, 1930; Am. J. Trop. Med. **13**:485, 1933.
- Johnson, H. W., and Hamann, E. E.: Am. J. Pub. Health **23**:917, 1933.
- Hughes, M. L.: Lancet **2**:238, 1896; Mediterranean, Malta or Undulant Fever, New York, The Macmillan Company, 1897.
- Hull, T. G., and Black, L. A.: J. A. M. A. **88**:463, 1927.
- Jenkins, P. K.: J. A. M. A. **92**:1593, 1929.
- Jordan, C. F.: J. Infect. Dis. **48**:526, 1931.
- Kampmeier, R. H.: Am. J. M. Sc. **176**:177, 1928.
- Keefer, C. S.: Bull. Johns Hopkins Hosp. **35**:6, 1924.
- Kennedy, J. C.: J. Roy. Army M. Corps **15**:317, 1910; **22**:9, 1914.
- Kern, R. A.: Am. J. M. Sc. **176**:405, 1928.
- King, W. F.: J. A. M. A. **91**:552, 1928.
- Kohlbry, C. O.: Minnesota Med. **12**:414, 1929.
- de Korte, W. E.: South African M. Rec. **22**:478, 1924.
- Kristensen, M.: Ugesk. f. læger **90**:869, 1928.
- Helms, T., and Martensson, A.: Ugesk. f. læger **93**:51, 1931.
- Kulowski, J., and Vinke, T. H.: J. A. M. A. **99**:1656, 1932.
- Lake, G. C.: Pub. Health Rep. **37**:2895, 1922.
- Larsen, W. P., and Sedgwick, J. P.: Am. J. Dis. Child. **6**:326, 1913.
- Leavell, H. R.; Poston, M., and Amoss, H. L.: Arch. Int. Med. **48**:1186, 1931.
- Lehmann, K. B., and Neumann, R. O.: Determinative Bacteriology, New York, G. E. Stechert & Company, 1931, vol. 2.
- Levin, W.: J. Lab. & Clin. Med. **16**:275, 1930.
- Lewis, S. J.: New Orleans M. & S. J. **83**:26, 1930.
- Loewy, I. D.: U. S. Vet. Bur. M. Bull. **6**:635, 1930.
- Löffler, W.: Schweiz. med. Wchnschr. **59**:304, 1929.
- McAlpine, J. G., and Mickle, F.: Am. J. Pub. Health **18**:609, 1928.
- and Slanetz, C. A.: J. Infect. Dis. **42**:66 and 73, 1928.
- McCulloch, T.; Weir, J. C., and Clayton, F. H. A.: Great Britain Mediterranean Fever Commission Reports, London, Harrison & Sons, 1907, pt. 7, pp. 74 and 218.
- McFadyean, J., and Stockman, S.: J. Comp. Path. & Therap. **22**:264, 1909.
- MacNeal, W. J., and Kerr, J. E.: J. Infect. Dis. **7**:469, 1910.
- Manson-Bahr, P.: Lancet **1**:1178, 1933.
- and Willoughby, H.: Brit. M. J. **1**:633, 1929.
- Marr, D. M.: Brit. M. J. **1**:959, 1933.
- Marston, J. A.: Great Britain Army Medical Reports of 1861, p. 486.

- Mason, C. F.: New York M. J. **78**:267, 1903.  
Mason, E. M.: J. M. A. Alabama **1**:50, 1931.  
Melvin, A. D.: U. S. Dept. Agric., Bur. Animal Ind., circ. 198, 1912.  
Meyer, K. F., and Eddie, B.: Proc. Soc. Exper. Biol. & Med. **27**:222, 1929.  
— and Shaw, E. B.: J. Infect. Dis. **27**:173, 1920.  
Miller, S.: Lancet **1**:1177, 1933.  
Mohler, J. R., and Eichhorn, A.: J. A. M. A. **58**:1107, 1912.  
— and Traum, J.: Ann. Rep. Bur. Animal Ind., 1911, p. 147.  
Moorehead, M. T.: M. Bull. Vet. Admin. **8**:195, 1932.  
Morales-Otero, P.: Puerto Rico J. Pub. Health & Trop. Med. **5**:144, 1929; J. Infect. Dis. **52**:54, 1933.  
— and Monge, G.: Puerto Rico J. Pub. Health & Trop. Med. **8**:193, 1932.  
Muir, Robert: A Textbook of Pathology, New York, Longmans, Green & Co., 1931, p. 440.  
Müller, L. R.: München. med. Wchnschr. **78**:1813, 1931.  
Nyfeldt, A.: Ugesk. f. læger **92**:491, 1930.  
O'Neil, A. E.: Ohio State M. J. **29**:438, 1933.  
Orpen, L. J. J.: South African M. Rec. **21**:325, 1923.  
Owen, S. A., and Newham, H. B.: Lancet **2**:536, 1915.  
Phease, R. N.: J. Roy. Army M. Corps **61**:296, 1933.  
Rainsford, S. G.: Irish J. M. Sc. **88**:150, 1933.  
Rawak, F., and Braun, R.: Klin. Wchnschr. **10**:776, 1931.  
Rodriguez de Parfearroyo, F.: Siglo méd. **76**:53, 1925.  
Rössle, R.: München. med. Wchnschr. **80**:5, 1933.  
Roger, H.: Marseille-méd. **68**:727, 1931.  
Ross, E. H.: J. Trop. Med. **9**:17, 1906.  
Ross, G. R.: (a) J. Hyg. **26**:403, 1927; (b) Tr. Roy. Soc. Trop. Med. & Hyg. **21**:57, 1927.  
Rothenberg, R. C.: Ann. Int. Med. **6**:1275, 1933.  
Samut, R.: Lancet **2**:878, 1911.  
Sander, J. F.: J. Michigan M. Soc. **32**:109, 1933.  
Sasano, K. T.; Caldwell, D., and Medlar, E. M.: J. Infect. Dis. **48**:576, 1931.  
Schilling, G. S.; Magee, C. F., and Leitch, F. M.: J. A. M. A. **96**:1945, 1931.  
Schlesmann, C.: Klin. Wchnschr. **11**:1711, 1932.  
Schottmüller, H.: Deutsche med. Wchnschr. **56**:1813, 1930.  
Schroeder, E. C., and Cotton, W. E.: Am. Vet. Rev. **40**:195, 1911.  
Scott, R. W., and Saphir, O.: Am. J. M. Sc. **175**:66, 1928.  
Sedgwick, J. P., and Larsen, W. P.: Am. J. Dis. Child. **10**:197, 1915.  
Sensénich, R. L., and Giordano, A. S.: J. A. M. A. **90**:1782, 1928.  
Sargent, E.; Gillot, V., and Lemaire, G.: Ann. Inst. Pasteur **22**:209, 1908.  
Simpson, W. M.: Ann. Int. Med. **4**:238, 1930; South. Surgeon **1**:184, 1932.  
— and Fraizer, E.: J. A. M. A. **93**:1958, 1929.  
Smith, T.: J. Exper. Med. **29**:451, 1919; **43**:207, 1926; Medecine **8**:193, 1929.  
Starr, L. E., and Maxcy, K. F.: Virginia M. Monthly **60**:218, 1933.  
Stone, C. T.: Texas State J. Med. **25**:225, 1929.  
Strong, R. P.: Undulant Fever, in Nelson Loose-Leaf Living Medicine, New York, Thomas Nelson & Sons, 1931, vol. 2, p. 209.  
— and Musgrave, W. E.: Philadelphia M. J. **6**:996, 1909.  
Tappau, J. W.: Texas State J. Med. **19**:176, 1923.

- Thompson, A.: Irish J. M. Sc. **72**:655, 1931.  
Thompson, R.: Canad. M. A. J. **29**:9, 1933.  
Tilghman, S. J.: Delaware State M. J. **5**:156, 1933.  
Topley, W. W. C., and Wilson, G. S.: The Principles of Bacteriology and Immunity, New York, William Wood & Company, 1929, (a) p. 1116; (b) p. 509.  
Trenti, E.: Policlinico (sez. med.) **30**:1249, 1923; ibid. (sez. prat.) **32**:767, 1925.  
Trotta, G.: Wien. klin. Wchnschr. **26**:1395, 1913.  
Tyndale, W. R., and Viko, L. E.: J. A. M. A. **81**:1953, 1923.  
Vanni, V.: Riforma med. **41**:555, 1925.  
Wade, E.: Lancet **1**:1342, 1933.  
Wainwright, C. W.: (a) Bull. Johns Hopkins Hosp. **45**:133, 1929; (b) South. M. J. **22**:1049, 1929.  
Watkins, W. W., and Lake, G. C.: J. A. M. A. **89**:1581, 1927.  
Weigmann, F.: Arch. f. Hyg. **102**:77, 1929.  
Weil, S.: Zentralbl. f. Chir. **57**:1269, 1930.  
Wellman, C.; Eustis, A., and Schochet, S. S.: Am. J. Trop. Dis. **1**:393, 1913.  
Whitehouse, B.: Brit. M. J. **2**:1095, 1929.  
Widal, F.; Léon-Kindberg, and Coton: Bull. Acad. de méd., Paris **64**:328, 1910.  
Williams, E. M.: J. Roy. Army M. Corps **9**:59, 1907.  
Wilson, G. S.: Brit. M. J. **2**:679, 1930; Bull. Hyg. **6**:389, 1931.  
Winslow, C. E. A.; Broadhurst, J.; Buchanan, R. E.; Krumwiede, C.; Rogers, L. A., and Smith, G. H.: J. Bact. **5**:191, 1920.  
Wohlfwill, F.: Virchows Arch. f. path. Anat. **286**:141, 1932.  
Wright, A. E., and Semple, D.: Brit. M. J. **1**:1214, 1897.  
— and Smith, F.: Lancet **1**:656, 1897.  
Yeckel, H. C., and Chapman, O. D.: J. A. M. A. **100**:1855, 1933.  
Yount, C. E., and Looney, R. N.: Arizona M. J. **1**:18, 1913.

## News and Notes

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**University News, Promotions, Resignations, Appointments, Deaths, etc.**—Ulrik Quensel, emeritus professor of pathology in the University of Uppsala, Sweden, has died at the age of 71.

The knighthood of the Order of the British Empire has been conferred on Frederick G. Banting of the University of Toronto.

M. G. Ramon, chief of the veterinary service of the Pasteur Institute, has been appointed subdirector of the institute. Dr. Ramon succeeds to the chair at the Academy of Medicine occupied by the late Emile Roux.

Hans Zinsser will spend the second half of the academic year 1934-1935 as an exchange professor at the University of Paris from Harvard University.

John A. Kolmer has been elected director of the Research Institute of Cutaneous Medicine in Philadelphia.

Louis Martin, chief of the serotherapeutic service of the Pasteur Institute in Paris, has been appointed director of the institute. Dr. Martin has been associated with the institute for forty years.

William Snow Miller, emeritus professor of anatomy at the University of Wisconsin, received the Trudeau medal at the thirtieth annual meeting of the National Tuberculosis Association in Cincinnati.

**Journal of the Mount Sinai Hospital, New York.**—This journal will be devoted principally to case reports. From time to time formal annual lectures delivered at the hospital may be included. The first number has been published.

**Mary Putnam Jacoby Fellowship.**—The Women's Medical Association of New York City announces that this fellowship (\$1,000 for one year) is open for investigative work in the medical sciences to women who are graduates of approved medical schools. Applications for 1934-1935 should be filed with Dr. Rose Cohen, 36 West Ninetieth Street, New York, not later than Sept. 1, 1934. With the applications should go statements as to health, educational qualifications and problems for investigation.

**Library of Legal Medicine.**—According to the Harvard Alumni Bulletin there has been established in the department of legal medicine of the Harvard Medical School a library of legal medicine. The department and library owe their existence to the generosity of Mrs. Frances Glessner Lee and bear the name of George Burgess Magrath, the present professor of legal medicine and a medical examiner for Suffolk County, Boston.

**Medals of Scientific Exhibit.**—The gold medal of the Scientific Exhibit of the American Medical Association at its last meeting, in Cleveland, was awarded to Gregory Shwartzman, Mount Sinai Hospital, New York, for his original investigations of skin reactivity to bacterial filtrates, its rôle in immunology and its practical applications. The silver medal was awarded to Timothy Leary, medical examiner for Suffolk County, Boston, for original work on the relation of cholesterol to atherosclerosis.

**Society News.**—The American College of Physicians will meet in Philadelphia on April 29 to May 3, 1935.

The ninth congress of the Far Eastern Association of Tropical Medicine will be held in Nanking on Oct. 1 to 7, 1934. The secretary of the executive committee is Dr. P. Z. King, National Health Administration, Nanking, China.

The Neisserian Medical Society of Massachusetts is composed of some seventy physicians who are interested especially in gonorrhea. The official organ of the society is the *New England Journal of Medicine*. The object of the society is to give members of the medical profession the benefit of the experience of the specialist in gonococcic infection.

# Abstracts from Current Literature

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## Pathologic Anatomy

CONGENITAL ANOMALIES OF THE CORONARY ARTERIES WITH CARDIAC HYPER-TROPHY. E. F. BLAND, P. D. WHITE and J. GARLAND, Am. Heart J. 8:787, 1933.

A male infant who died at the age of 3 months showed an abnormal origin of the left coronary artery from the pulmonary artery, associated with marked enlargement of the heart (due to hypertrophy and dilatation of the left ventricle) and extensive degenerative changes in the ventricular wall supplied by the malposed vessel. In view of these findings it is probable that the paroxysmal attacks of acute discomfort precipitated by exertion and associated with profound vasomotor collapse occurring in this infant were those of angina pectoris. The electrocardiographic picture was similar to that of an adult with severe coronary disease. In the few recorded cases of this rare anomaly (eight in addition to the one reported in this article) a characteristic pathologic picture has resulted. Death within the first year has been the rule. Two of the cases have been exceptional.

### AUTHORS' SUMMARY.

ANOMALOUS ORIGIN OF THE LEFT CIRCUMFLEX CORONARY ARTERY. WILLIAM ANTOPOL and M. A. KUGEL, Am. Heart J. 8:802, 1933.

Four hearts were studied in which there were an anomalous origin and course of the left circumflex coronary artery. In each heart the left anterior descending branch and the right coronary artery had their normal origin and distribution. In three hearts the left circumflex coronary artery arose directly from the right sinus of Valsalva, immediately posterior to the origin of the right coronary artery. It then pursued its course posteriorly to the root of the aorta and the left auricular appendage around the margo obtusus in the auriculoventricular sulcus. In the fourth case the left circumflex coronary artery arose as a branch of the right coronary artery 1 cm. from its ostium. It then maintained a course similar to that described for the left circumflex coronary artery in the other three cases. A point of clinical interest in one of these cases is the fact that the left circumflex coronary artery was normal and patent throughout, whereas the right coronary artery and the left anterior descending branch presented numerous occlusions. The clinical history of the 51 year old patient gave ample evidence of repeated attacks of coronary occlusion in the last three years of his life. It seems possible that the independent origin and distribution of the left circumflex coronary artery in this case served for a time as a compensating source of nutrition to the myocardium through its anastomoses.

### AUTHORS' SUMMARY.

LUNGS AFTER TREATMENT OF ASPHYXIA NEONATORUM IN THE DRINKER RESPIRATOR. D. P. MURPHY and J. T. BAUER, Am. J. Dis. Child. 45:1196, 1933.

The pathologic changes in the lungs of thirty asphyxiated infants who were given artificial respiration in the Drinker respirator, but who died within twenty-four hours after birth, are compared with those in a similar number who had not received this means of artificial respiration. Seventy per cent of the thirty deaths were the result of either intracranial injury or prematurity. No death was due to a pulmonary condition. A slight increase in the incidence of pulmonary congestion followed the use of the Drinker respirator; this indicated that the treatment had an appreciable effect on the contents of the chest. Artificial respiration had no influence on the kind of cellular elements observed in the air passages, but

may have drawn amniotic débris from the bronchioles to the alveoli. Otherwise, no gross or microscopic changes were recognized in the lungs of the treated infants that had not been seen in the lungs of untreated infants.

## AUTHORS' SUMMARY.

"DOUBLE APPENDIX" ASSOCIATED WITH OTHER CONGENITAL ANOMALIES. HENRY N. PRATT, Am. J. Dis. Child. **45**:1263, 1933.

Because of an imperforate anus, sigmoidostomy was performed on a boy born at full term, who died fifteen days later, after a course of septic fever. Necropsy revealed mesenteric attachments of the midline embryonic type. The rectum was congenitally absent, while the sigmoid ended in a blind pouch at the base of the posterior aspect of the bladder. The sigmoid was represented by one small loop of fused bowel with a continuous lumen. Above the pouchlike sigmoid was about 7 cm. of intestinal tract, presumably colon, stretching upward and somewhat to the right. At this point there were two "appendixes" on opposite sides of a slight fusiform swelling, presumably the cecum, each having an individual mesenteric attachment to the ileum. Two lateral taeniae on the colon terminated at the bases of the "appendixes." The ileum was directly continuous with, and had practically the same caliber as, the preceding colon. Above the colon the enteric tract appeared to be normal. Pratt points out that the anomalous character of the enteric tract in this case is suggestive of avian morphology.

RALPH FULLER.

ATELECTASIS OF THE NEW-BORN. SIDNEY FARBER and JAMES L. WILSON, Am. J. Dis. Child. **46**:572, 1933.

A certain degree of initial atelectasis is physiologic for probably several days after birth. In this type of unexpanded lung the alveoli are small, circular and lined with cuboid epithelium. In the lungs of infants who have lived several days areas of resorption atelectasis may be demonstrable. In these areas the collapsed alveoli present tortuous walls and are lined by flattened cells. In premature infants there may be solid areas of pulmonary parenchyma superficially resembling atelectasis but explained by incomplete development or immaturity of these areas.

RALPH FULLER.

ENDOMETRIOSIS OF LYMPH NODES. G. H. HANSMANN and J. R. SCHENKEN, Am. J. Obst. & Gynec. **25**:572, 1933.

In two cases endometrial tissue was found in the regional lymph nodes of the uterus post mortem. One of the patients had had a syncytoma malignum in which endometrial tissue may have entered the lymphatics. The vessels of the endometrium are opened during menstruation. Viable endometrial tissue has been found free in the oviducts as well as in the venous sinuses and the lymphatic vessels of the uterus.

JACOB KLEIN.

THE MICROINCINERATION OF HERPETIC INTRANUCLEAR INCLUSIONS. L. E. RECTOR and E. J. RECTOR, Am. J. Path. **9**:587, 1933.

Microscopic examination of incinerated herpetic intranuclear inclusions from the cerebral cortex of rabbits reveals the presence of considerable inorganic material in young full inclusions with a progressive decrease in amount as the inclusions develop. Mature inclusions are frequently devoid of any inorganic residue.

## AUTHORS' SUMMARY.

RARE FORM OF SACCULAR CARDIAC ANEURYSM WITH SPONTANEOUS RUPTURE. W. C. HUNTER and R. L. BENSON, Am. J. Path. **9**:593, 1933.

A man, 45 years old, died suddenly of spontaneous rupture of the thin fibrous wall of an aneurysm of the external aspect of the left ventricle. The wall of the

aneurysm varied between 0.1 and 0.7 cm. in thickness; the aneurysm appeared to have originated in a small area of myocardial fibrosis.

CONTROLLED FORMATION OF COLLAGEN AND RETICULUM. S. BURT WOLBACH,  
Am. J. Path. 9:689, 1933.

Fibrin and other preformed materials do not contribute to collagen formation in repair by organization. Collagen and reticulum are physical variations of the same material. Collagen is the product of the secretory activity of fibroblasts, and its alinement and distribution are determined by the shape of the cell and its processes, including fibroglia fibrils.

AUTHOR'S CONCLUSIONS.

THE HISTOPATHOLOGY OF THERAPEUTIC (TERTIAN) MALARIA. WALTER L.  
BRUETSCH, Am. J. Psychiat. 12:19, 1932.

The immediate tissue reaction of the body to the malarial plasmodium consists in a stimulation of the reticulo-endothelial apparatus (system of histiocytes), leading to a new formation of macrophagic tissue in various organs. Both the specific endothelia of the liver, spleen, lymph nodes and bone marrow and the histiocytes of the connective tissue take part in the stimulation. In therapeutic malaria the histiocytes of the blood are mainly derivatives of the specific endothelium. To a minor degree, common capillary endothelium is engaged in the formation of intravascular endothelial phagocytes. This has been established for the endothelium of the capillaries of the cortex of the brain and for the endothelial cells of the capillary venules of certain connective tissues. By means of the supravital technic, it has been found that the intravascular macrophagic phagocytes are clasmacytotes in the sense of Sabin, Doan and Cunningham. Although the capillary endothelial cells show signs of stimulation, they do not become phagocytic while they retain their anatomic position in the walls of the vessels.

In addition to involvement of the histiocytes, there is an activation of the undifferentiated embryonic mesenchymal cells. The fibroblast, the mesothelial cell and the histiocyte are distinct types of cells. While both the fibroblast and the mesothelial cell are also capable of stimulation, they can be distinguished from the active histiocyte in malaria-infected tissue.

In the nervous system the macrophagic response has been greatest in the lepto-meninges. In the arachnoid the malaria-stimulated histiocytes stand out distinctly from the less active arachnoid lining cell. In the adventitial sheaths of the vessels of the cortex of the brain the mesodermal phagocytes are only slightly stimulated. A small increase in the number of macrophages has been found about middle-sized and large cortical vessels. Stimulated histiocytes are more numerous in the perivascular spaces of the large vessels in the white matter, and in the striatum and pons. The small mesodermal elements along the capillaries of the cortex of the brain have not been seen to be activated. The microglia, as a whole, do not take part in the general reaction of the reticulo-endothelial system.

Therapeutic malaria produces an activation of the mesodermal tissue in which stimulation of the histiocytes and activation of the undifferentiated mesenchymal cells are outstanding features.

AUTHOR'S SUMMARY.

THE PATHOLOGICAL ANATOMY OF PULMONARY TUBERCULOSIS IN THE AMERICAN NEGRO AND IN THE WHITE RACE. FRANKLIN R. EVERETT, Am. Rev. Tuberc. 27:411, 1933.

The anatomic characteristics of tuberculosis differ widely in American Negroes and white persons of the same community in that the disease pursues a more rapidly fatal course in the Negroes and more frequently takes the form of a widespread pneumonia with rapid excavation of the lung and less conspicuous formation of fibrous tissue. The type of pulmonary tuberculosis which is prevalent in adult white persons occurs in only half of the Negroes who contract pulmonary tuber-

culosis and pursues in them a more rapid course. The type of pulmonary tuberculosis characteristic of childhood, with caseous tracheobronchial lymph nodes, occurs in nearly 50 per cent of adult Negroes, but in only a small percentage of adult white persons who die of pulmonary tuberculosis. In more than half of the adult Negroes with the childhood type of pulmonary tuberculosis the lesion has its origin in the apex of the lung. Latent apical tuberculosis occurs considerably less frequently in adult Negroes than in adult white persons.

H. J. CORPER.

FATTY INFILTRATION OF THE MYOCARDIUM. O. SAPHIR and M. CORRIGAN, Arch. Int. Med. 52:410, 1933.

By fatty infiltration of the myocardium is meant the formation of an abundance of subepicardial fat which extends into the myocardium and occupies the usual position of the muscle fibers. A short review of the literature on fatty infiltration of the myocardium is given, and a study of fifty-eight cases is reported. The myocardium of the right ventricle is the region mainly affected. The infiltration leads to a replacement of the muscle fibers by fatty tissue. The muscle fibers primarily become atrophic and later apparently disappear. If the replacement by fat involves a large portion of the myocardium it may lead to sudden death. Only if careful autopsy reveals the absence of all other major lesions may fatty infiltration of the myocardium be regarded as the sole cause of death.

Two cases of this type are reported. In two other cases replacement of the myocardium by fat was in all probability the cause of death. In one the diagnosis was pseudohypertrophic muscular dystrophy in which the fatty infiltration of the myocardium was probably a part of the general replacement of muscle with fat. In the second the patient died after partial thyroidectomy. In twenty-nine instances other pathologic changes, in addition to fatty infiltration of the myocardium, were found at autopsy. In these cases the lesion in the heart was thought to be a factor in hastening death. In twenty-five instances fatty infiltration of the myocardium was an incidental finding at autopsy. A clinical study of the patients whose cases are reported reveals that fatty infiltration of the myocardium may cause death without any premonitory symptoms of heart failure. When such infiltration is present, factors which cause an increased demand on the heart and which under normal conditions could easily be compensated for may lead to sudden death. Fatty infiltration of the myocardium may be regarded as a morphologically demonstrable cause of heart failure and death in instances in which death clinically was thought to have been the result of functional disorders without a morphologic basis. Further study with exact clinical methods might establish a complex of signs and symptoms sufficiently characteristic to warrant a clinical diagnosis of fatty infiltration of the myocardium.

AUTHORS' SUMMARY.

THROMBOSIS OF INTRACRANIAL ARTERIES. H. H. HYLAND, Arch. Neurol. & Psychiat. 30:342, 1933.

Hyland records three cases in which the clinical and pathologic features were peculiar. In the first, necropsy revealed thrombosis of both anterior cerebral arteries which was clinically manifested by complete paralysis of the left leg, partial paralysis of the left arm, apraxia and a grasp reflex. There were also mental disturbances—euphoria, freedom from inhibitions and motor activity in the nonparalyzed extremities ("the arm, for instance, was in a constant state of purposeful activity, grasping at anything that came within reach"). In the second case there were sudden weakness in the left extremities, hypotonia, active tendon reflexes, diplopia and dysphagia. The pathologic diagnosis was: thrombosis of the left vertebral and basilar arteries, cerebral arteriosclerosis and acute sphenoid sinusitis. No evidence of syphilis was found, though the Wassermann reaction was 4+. In case 3 the clinical signs were: sudden flaccid hemiplegia on the left side, blindness in the right eye and failure of its pupil to react to light, and sensory disturbances over the paralyzed face and arm. There were thrombosis of the

central artery of the retina and of the right carotid and middle cerebral arteries, softening of the parts of the brain supplied by the latter and demyelinization of the right optic nerve.

G. B. HASSIN.

FAMILIAL SPASTIC PARALYSIS. HARRY A. PASKIND and THEODORE T. STONE, Arch. Neurol. & Psychiat. 30:481, 1933.

Whereas spastic paraplegia involving the lower extremities only or both the lower and upper extremities, with or without bulbar and mental manifestations, has repeatedly been described from the clinical standpoint (in 152 families), histologic reports are exceedingly rare. The patient of Paskind and Stone was one of five siblings. Of these, two brothers were also afflicted with spastic paraplegia, and all three were idiots; one other brother and a sister, aged 9 months, were normal. The remarkable macroscopic features were: complete absence of sulci and convolutions over the parietal and occipital lobes except at the tips of the latter, numerous brown areas of gray matter within the white substance (heterotopia) and defective myelinization, especially in the white matter of the occipital and parietal lobes. The spinal cord showed no changes. In the parietal and occipital lobes the defective myelinization was associated with agyria—the structures regulating motion and acting as cortical receptors for sensation were not developed. The extrapyramidal fibers were normal. Absence of association fibers in the parietal and occipital lobes had resulted in pachygryria.

G. B. HASSIN.

ANEURYSM OF THE INTERNAL CAROTID ARTERY. ROBERT ZOLLINGER and ELLIOTT C. CUTLER, Arch. Neurol. & Psychiat. 30:607, 1933.

A patient presented symptoms and signs of involvement of the second, third, fourth, fifth, seventh and eighth cranial nerves on the right and an enlarged sella turcica, with destruction of the clinoid processes. The condition proved to be due to an aneurysm of the right carotid artery, which had eroded the sphenoid and temporal bones, infringed on the sella turcica, become adherent to the fourth and fifth nerves and displaced the other cranial nerves. The pituitary gland was not grossly identified and was apparently a part of the aneurysmal wall. The latter showed syphilitic arteritis.

G. B. HASSIN.

CHANGES IN THE BRAIN IN LEGAL ELECTROCUTION. GEORGE B. HASSIN, Arch. Neurol. & Psychiat. 30:1046, 1933.

Organic changes were found in the brains of five criminals executed by electric shocks. The parenchymal changes were: tears, fissuration and cracks of the brain tissue; dislocation, swelling and even liquefaction of the ganglion cells; demyelinization of the white fibers, and an enormous so-called swelling of the oligodendroglia, with dilatation of the shrinkage spaces of His and rupture of the vascular tunics, especially of the elastic membrane. The latter in all the cases was broken up and formed loops, and in some instances the muscularis and the adventitia were also fragmented. Hemorrhages were not present. Another remarkable change was the presence of reactive phenomena (satellitosis and neuronophagia) in the deeper layers of the cortex and in the subarachnoid space (cell proliferation), in spite of the short interval between the action of the electric current and the onset of death. Some changes were similar to those seen in concussion of the central nervous system or in conditions of increased intracranial pressure.

AUTHOR'S ABSTRACT.

THROMBOPHLEBITIS OF THE INFERIOR VENA CAVA AND OCCLUSION OF THE HEPATIC VEINS. R. H. RIGDON, Bull. Johns Hopkins Hosp. 53:162, 1933.

The literature on endophlebitis hepatica obliterans was reviewed to determine whether such a lesion ever exists in the absence of phlebitis of the hepatic portion of the inferior vena cava. A case is reported in which both lesions were associated.

In the majority of cases of endophlebitis hepatica obliterans reported in the literature the disease was associated with inflammation of the hepatic portion of the inferior vena cava, and there is little support for the opinion that the two processes are different. Furthermore, there appears to be little, if any, justification for the view that endophlebitis hepatica obliterans is a distinct entity.

AUTHOR'S SUMMARY.

**CONGENITAL CYSTIC DISEASE OF THE LUNGS.** D. H. COLLINS, J. Path. & Bact. **37:**123, 1933.

A case of congenital cystic disease of both lungs is described with a peculiar giant cell hyperplasia of unknown cause in all the lymph nodes.

**MASSIVE PARAVERTEBRAL HETEROTOPIA OF MARROW IN A CASE OF ACHOLURIC JAUNDICE.** S. J. HARTFALL and M. J. STEWART, J. Path. & Bact. **37:**455, 1933.

A case of massive heterotopia of bone marrow occurring in the thorax of a patient who died of familial acholuric jaundice is reported. The possible relationship of these two conditions is discussed.

AUTHORS' SUMMARY.

**THE ISLANDS OF LANGERHANS IN OBESITY.** R. F. OGILVIE, J. Path. & Bact. **37:**473, 1933.

A method is described whereby the percentage area of islet tissue in the pancreas and the average area of the islands can be estimated. By this method the islets of Langerhans in nineteen obese patients were compared with those in nineteen lean subjects. Of the obese group thirteen, or 68 per cent, were found to possess (1) an abnormally high percentage area of islet tissue, (2) a normal number of islands per given area and (3) islands the average size of which was definitely greater than the normal. The relationship between the hypertrophied condition of the islets and the obese state is discussed.

AUTHOR'S SUMMARY AND CONCLUSIONS.

**STRUCTURAL CHANGES IN ANTIRABIC TREATMENT PARALYSIS.** S. GETZOWA, G. STUART and K. S. KRIKORIAN, J. Path. & Bact. **37:**483, 1933.

The predominant feature in two cases of Landry's paralysis following antirabic treatment and in one case of unknown origin was a widespread lesion of ganglion cells inducing rapidly advancing fatal paralysis. The total absence of perivascular zones of demyelination and perivascular cuffing in the central nervous system argues against the inclusion of antirabic treatment among the somewhat heterogeneous group of factors (smallpox, vaccinia, measles, varicella and typhoid fever) capable of producing acute disseminated encephalomyelitis (Westphal). It also argues against the theory of a virus generally.

**MORBID ANATOMY AND HISTOLOGY OF ASBESTOSIS.** S. ROODHOUSE GLOYNE, Tubercl **14:**445, 493 and 550, 1933.

The pathologic result of the inhalation of asbestos dust is different from that of the inhalation of other dusts that give rise to diseases, owing to the peculiar long needle-like shape of the particles of asbestos. The fibrosis is diffuse, and nodules such as characterize silicosis are not seen. The various conditions in the asbestos industry render it probable that as time goes on unusual forms of asbestosis will occur. In the pleura the disease is obliterative. In an uncomplicated case the lung shows dense bluish-black areas corresponding to the secondary lobules, surrounded by thick bands of interlobular connective tissue, generally with a reddened background of terminal bronchopneumonia in the less affected portions of the lung. In other organs signs of the disease visible to the naked eye are few.

The affected lung shows particles of five different kinds: (a) the carbon pigment common to all dwellers in towns; (b) an amorphous brown pigment, presumably blood; (c) sharp, jagged particles, probably carbonaceous; (d) fibers of asbestos, and (e) asbestosis bodies. The fibers of asbestos can also be found readily in the upper respiratory tract. The tissue reaction to the fibers is threefold: (a) thickening of the fibers to form asbestosis bodies; (b) cellular changes, chiefly accumulation of large phagocytic cells containing the dust and formation of giant cells, and (c) fibrosis of the type common to all forms of pneumonoconiosis. The giant cell of asbestosis is a minute collection of phagocytes. The immobilization and long persistence of the phagocytes and especially of the giant cells are characteristic.

The most marked histologic feature of the lung is the holding up of the fibers at the distal ends of the respiratory bronchioles and in the alveolar ducts with the accumulation of large mononuclear phagocytes and the giant cells. In the later stages of the disease, the lymphatics and the adjacent air sacs became filled with asbestos dust and phagocytes. Finally there occurs an increase of connective tissue around the bronchioles, alveolar ducts, air sacs, capillaries and venules, in the interlobular septums and beneath the pleura. These changes ultimately result in complete obliteration of all pulmonary configuration. The disease tends to be most marked in the lower lobes; adherent pleura is almost the rule; the long strand-like or bandlike adhesions seen so frequently in tuberculosis have not been encountered. Pleural effusion is rare. The asbestosis body is a regular concomitant of the disease, but structures closely resembling it are found in other pneumonoconioses. In case of doubt, the golden yellow pigment should be dissolved with strong sulphuric acid. The central fiber of asbestos thus displayed is usually different from the particles seen in pseudo-asbestosis bodies. Tuberculosis and bronchopneumonia are commonly associated with asbestosis, as they are also with silicosis.

H. J. CORPER.

**NECROSIS IN THE LIVER FROM POISONING WITH THYROXINE. F. GERLEI,**  
*Ann. d'anat. path. 10:555, 1933.*

The toxicity of thyroxine was studied in rabbits. Daily injection of 4 mg. of thyroxine under the skin resulted in the death of the animals within from six to seven days, with extreme emaciation. Central necrosis of the hepatic lobules was found.

AUTHOR'S SUMMARY.

**SITE OF FORMATION OF THE SEX HORMONE IN THE HYPOPHYSIS. E. J. KRAUS,**  
*Beitr. z. path. Anat. u. z. allg. Path. 91:245, 1933.*

Granting that the hormone which motivates maturation of the ovarian follicle and which is excreted in the urine of pregnant women is produced by the hypophysis, Kraus briefly reviews the evidence favoring the formation of the hormone by one or the other of the three types of cells of the hypophysis. He then presents the results of his histologic study of the hypertrophied hypophyses of seven nonpregnant women with carcinoma of the genital tract and of two women and five men with tumor of the brain. At necropsy urine from the bladder of four of the patients with carcinoma and of three of those with tumor of the brain gave a positive reaction for the hormone by the mouse test. Kraus interprets his findings as indicative of the formation of the hormone by the eosinophilous cells of the anterior lobe. While the mere weight of evidence may be so interpreted, it is to be noted that an individual case may speak just as strongly in favor of formation by one of the other two types of cells. He next presents the results of a series of implantations of bits of the normal and the adenomatous human hypophysis into immature mice. These results, Kraus thinks, indicate that the hormone may be formed by either type of chromophil. Determination of the source of the hormone in pregnancy requires further research.

O. T. SCHULTZ.

MALFORMATIONS OF THE LOWER PART OF THE BODY. E. NACHMANSOHN, Frankfurt. *Ztschr. f. Path.* **44**:117, 1932.

A stillborn child is described, measuring 36 cm. In place of the lower extremities there was a tail-like structure, 10 cm. in length, taking origin from the pelvic region and becoming gradually smaller to end in a finger-like projection. The external genital organs and the anus were missing. In the region of the lowest portion of the lumbar segment was a small soft projection 1.4 cm. in length, which did not reveal an opening. Only one umbilical artery could be found. No urinary bladder, urachus or persistent cloaca could be made out. The lower portions of the abdominal muscle were markedly hypoplastic. The large intestines were distended and filled with meconium, and the sigmoid colon ended blindly. The surfaces of the serosa were smooth and glistening. Both suprarenal glands were present and normally located, but the kidneys were absent. Close to the umbilical artery two gonads were found, which resembled testes. There was also an occult spina bifida. The explanations for these malformations are given. The author recommends the following classification of malformations of the lower part of the body: (a) isolated defects of the external form of the caudal portion of the body, (b) isolated defects of the anlagen of the inner organs of the caudal end of the body, and (c) isolated defects of the external form of the caudal portion of the body (1) with normally formed and developed inner organs and (2) with defects of the inner organs. This classification does not seem justifiable because (a) and (1) overlap.

O. SAPHIR.

SYPHILIS OF THE JOINTS. E. FREUND, *Virchows Arch. f. path. Anat.* **289**:575, 1933.

This contribution from Erdheim's laboratory is based on the same material that formed the basis of the study of syphilis of bone reported in the preceding volume of *Virchows Archiv* (abstr., *ARCH. PATH.* **17**:587, 1934). Thirteen syphilitic joints were studied macroscopically, roentgenologically and microscopically; seven were from a single patient, four from another patient, and two from two other patients. In seven instances the involvement of the joint was secondary to perforation by a gumma of the epiphysis into the joint; the perforation usually occurred at the margin of the joint. Fibrous ankylosis was frequent, but cartilaginous union was seen only once. In four instances fractures extended through the weakened bone into the joint. In those instances in which a gumma could not be found either in the epiphysis or in the synovial membrane, the changes in the joint were of the same character as when the arthritis was due to perforation by a gumma into the joint, but were of lesser degree. In no instance was a gumma which had developed originally in the cartilage the cause of syphilitic arthritis. The cartilage of the joint revealed a variety of alterations, which ended sometimes in destruction and sometimes in new formation. Changes similar to those in arthritis deformans, fractures of the bone forming the surfaces of the joints and small hernial outpouchings of the synovial membrane are held to be the results of the syphilitic arthritis.

O. T. SCHULTZ.

GENERALIZED DISEASE OF THE OSSEOUS SYSTEM IN CHILDHOOD. E. HÄSSLER and KRAUSPE, *Virchows Arch. f. path. Anat.* **290**:193, 1933.

A condition which was clinically diagnosed as aleukemic myelosis in a child, 27 months old, who presented, in addition, osteosclerosis and terminal anemia, is the subject of a comprehensive clinical and roentgenologic report by Hässler. For comparison he includes similar studies of a case of generalized sarcomatosis of the skeleton, with osteolysis and osteoporosis, two cases of osteitis fibrosa cystica and one of Albers-Schönberg's disease, with active rickets, scurvy and anemia. All these conditions occurred in infants. Krauspe follows with a detailed histologic study of the bones in the case of aleukemic myelosis and in that of Albers-Schönberg's disease. He interprets the first condition as primary hyperplastic

disease of the bone marrow, with reactive osteosclerosis, which in turn led to anemia. In the case of Albers-Schönberg's disease both the osseous system and the hematopoietic system were primarily involved. The case of marble disease described had some of the characteristics of von Jaksch's anemia. Krauspe gives also the results of a histologic study of the bones in two cases of lymphatic leukemia and in one case of myeloid leukemia in children. Although osteolytic and osteoporotic changes predominate in leukemia, there may be subperiosteal new formation of sclerotic bone.

O. T. SCHULTZ.

**NODULAR LINGUAL MYOLYSIS.** A. H. ROFFO, Ztschr. f. Krebsforsch. **39**:464, 1933.

Roffo describes a second case of the condition previously termed by him "nodular lingual myolysis." This case occurred in a woman 31 years of age. The lesion was nodular and histologically showed evidence of progressive granulation and disappearance of muscular fibrillae, terminating in complete vacuolar degeneration. The sarcolemma was not affected. There was no involvement of the lymph glands. The only suggestion of an inflammatory reaction was that of a slight hypertrophy of the adjoining mucosa.

H. E. EGGERS.

### Microbiology and Parasitology

**HERPES ENCEPHALITIS PROBLEM.** F. P. GAY and M. HOLDEN, J. Infect. Dis. **53**:287, 1933.

Additional evidence for the theory that epidemic encephalitis is due to a neurotropic strain of the virus of herpes simplex operative under peculiar conditions of susceptibility on the part of the patient is reported.

FROM AUTHORS' SUMMARY.

**EXPERIMENTAL PERTUSSIS.** H. MACDONALD and E. J. MACDONALD, J. Infect. Dis. **53**:328, 1933.

A filter-passing virus plays no rôle in the etiology of pertussis. The disease is caused by the bacillus of Bordet and Gengou. Active immunity is conferred by the injection of Bacillus pertussis vaccine. [The basis for this summary is experiment on human volunteers.]

AUTHORS' SUMMARY.

**RELAPSING FEVER IN CALIFORNIA.** G. E. COLEMAN, J. Infect. Dis. **53**:337, 1933.

Relapsing fever caused by three strains of spirochetes isolated in California has been studied in mice, and the disease has been found to be more severe than that caused by *S. novyi* and possibly than that produced by *S. duttoni*. Occasionally symptoms attributable to nerve lesions have been observed. The clinical course of an accidental infection in a laboratory worker is described. The exact locality in California where the infective organism in the case originated is uncertain. The blood of two adult guinea-pigs taken on the twenty-second and twenty-third days, respectively, after inoculation infected mice. Although the blood of the guinea-pigs was examined daily, no spirochetes were ever seen. The serum of the animals at this time showed no protective properties against infection in mice. With the California strains, in the absence of spontaneous agglutination, little if any difference in virulence was shown between blood taken during the primary attack and that taken during the first three relapses.

AUTHOR'S SUMMARY.

**BACTERIA IN THE FILTRABLE STATE IN BACTERIOPHAGE.** A. I. KENDALL and A. W. WALKER, J. Infect. Dis. **53**:355, 1933.

Bacteriophage causes bacteria to disappear. Viable bacteria in the filtrable state are present in the filtrates of the phaged bacteria. These bacteria in the

filtrable state are invisible, unstainable and uncultivable by ordinary methods. Hitherto bacteria in the filtrable state have not been demonstrated consistently in filtered phage solutions. This is due presumably in part to difficulties in curbing the action of the phage and partly to circumstances attending the return of these bacteria in the filtrable state to the visible, stainable, nonfiltrable state in which they grow readily in ordinary mediums. Certain procedures are described which by curbing the action of the phage permit these invisible bacteria in the filtrable state to redevelop into nonfiltrable organisms. The methods employed for this purpose were: exposure to methylthionine chloride and sunlight; the addition of living homologous organisms, filtration and the subsequent recovery of the bacteria in the nonfiltrable state from the filtered clear phage solution; the addition of phage to K medium; the addition of killed cultures of the homologous organism; contact with specific antiphage; the use of bile. These procedures, admittedly far from ideal, have, in spite of their obvious imperfection, yielded positive results.

## AUTHORS' SUMMARY.

NORMAL FLORA OF PREPUBERTAL VAGINA. H. PETTIT and C. H. HITCHCOCK, J. Infect. Dis. 53:372, 1933.

Studies of the prepubertal vagina in sixty girls, most of whom were orthopedic patients, show that the flora is predominantly diphtheroid in character. Gram-positive cocci, such as nonhemolytic streptococci and nonpigment-forming staphylococci, are present less frequently and in smaller numbers. Organisms of the intestinal flora seldom if ever occur in healthy children, though in severely ill patients there is a distinct tendency for them, together with *Streptococcus aureus*, to overgrow the normally occurring flora.

## AUTHORS' SUMMARY.

EFFECTS OF COLLOIDAL SILICA ON EXPERIMENTAL TUBERCULOSIS IN GUINEA-PIGS. S. L. CUMMINS and C. WEATHERALL, J. Hyg. 33:295, 1933.

The experiments reported confirm the observation by Gye and Kettle that the addition of silica sol to tubercle bacilli leads, on inoculation into guinea-pigs, to a marked increase of local reaction and to a greater local pullulation of tubercle bacilli within the first few days after injection. While a shortening of the survival period was observed in the animals given tubercle bacilli along with silica sol as compared with the guinea-pigs infected with tubercle bacilli alone, the difference was slight and hardly to be regarded as significant. The adjuvant action of the silica sol seems to be local and transitory, as is the local irritant action of silica sol alone; and any tendency to a more rapid dissemination of bacilli appears to be neutralized by the local fibrosis which undoubtedly follows the introduction of silica sol into the tissues. These findings are of interest because they fall into line with the known fact that the tuberculosis death rate in industries involving exposure to silica dust is high in late middle age and after many years of dust inhalation, not in the early years of exposure as should be the case if the silica determined an early generalization of tuberculous infection.

## AUTHORS' SUMMARY.

STATISTICS OF ERYSIPelas IN ENGLAND AND WALES. W. T. RUSSELL, J. Hyg. 33:421, 1933.

The annual number of notified cases of erysipelas is approximately 17,000, and assuming complete notification of the disease the fatality rate in the cases is approximately 6 per cent. The death rate in terms of the population varies according to age, being highest at the beginning and the end of life and minimal between the ages of 5 and 10 years. The mortality of males is identical with that of females up to the age 25, but is afterward in excess. The disease has in recent years a well marked seasonal incidence—a winter and spring excess with a summer decrease. In this respect it resembles scarlet and puerperal fevers. Although

its seasonal incidence has changed in the course of time, the alteration has not been nearly so pronounced as that of scarlet fever. The incidence is highly correlated with overcrowded conditions. . . . The morbidity from erysipelas is fairly well correlated in time with that from scarlet fever and erysipelas, but in London and other urban districts and in the rural districts of England and Wales the special correlation is very small. Coefficient figures are given.

FROM AUTHOR'S SUMMARY.

**ANAEROBIC METHODS FOR THE IDENTIFICATION OF HAEMOLYTIC STREPTOCOCCI.**

R. M. FRY, J. Path. & Bact. **37**:337, 1933.

This article emphasizes the value of anaerobic cultures in routine efforts to isolate hemolytic streptococci.

**THE SIZE OF THE VIRUS OF LOUPING-ILL OF SHEEP.** W. J. ELFORD and I. A. GALLOWAY, J. Path. & Bact. **37**:381, 1933.

The size of the virus of louping-ill has been estimated to be from 15 to 20 microns by filtration through carefully graded collodion membranes. The filtration end-point has been checked by inoculation of both mice and sheep. Filtrates infective for mice also proved to be infective for sheep. The virus was found to have retained its infectivity for sheep after twenty-two passages in mice over a period of two hundred and ten days, and also after forty passages in mice extending over a period of six hundred and eighty-seven days. The virus quickly becomes inactivated when kept in broth at a room temperature of from 18 to 20 C. A suspension at  $p_{H_2} 7.6$ , initially infective in 1:100,000 dilution, had dropped 90 per cent in potency after twenty-four hours and was completely noninfective after three days. The virus may be stored satisfactorily for much longer periods at 4 C. Filtrates of broth suspensions at  $p_{H_2} 7.6$  and  $p_{H_2} 8.5$ , kept in small flasks closed with cotton-wool plugs, were found to be infective after seventy days. The virus exhibits greater stability in slightly alkaline broth, namely at  $p_{H_2} 7.5$  to 8.5. Infection has been successfully transmitted to mice with filtrates by a technic of intranasal instillation.

AUTHORS' SUMMARY.

**THE GROWTH PHASES OF PLEUROPNEUMONIA AND AGALACTIA ON LIQUID AND SOLID MEDIUM.** J. C. G. LEDINGHAM, J. Path. & Bact. **37**:393, 1933.

The morphology and growth phases of organisms isolated in cases of pleuropneumonias and agalactia have been studied in liquid and solid mediums, and the conclusion has been reached that these organisms may be placed, provisionally, in the family Actinomycetaceae. The question of the appropriate genus is reserved for further consideration.

AUTHOR'S SUMMARY.

**ACID-FAST ORGANISMS OTHER THAN MAMMALIAN TUBERCLE BACILLI FROM DISEASE IN MAN. AVIAN TUBERCLE BACILLI.** ARNOLD BRANCH, Tubercle **14**:337, 1933.

There are a number of authentic cases on record of acid-fast bacilli not mammalian tubercle bacilli infecting man. Some of these strains appear to belong to the avian group while others do not belong to any type of tubercle bacilli, but are probably new strains of pathogenic acid-fast bacteria. Particular difficulty is encountered in diagnosing infection with strains of avirulent avian tubercle bacilli, and a scheme is outlined by which these may be recognized. Avirulent avian tubercle bacilli are capable of forming tuberculin and of sensitizing fowls to a known tuberculin. Inoculation of the common white mouse has proved a useful aid in diagnosing infection with acid-fast strains which are not typical tubercle bacilli or saprophytes. After large intraperitoneal doses multiple abscesses develop in the kidneys.

H. J. CORPER.

PATHOLOGIC CHANGES IN LEUKOCYTES IN TUBERCULOSIS. M. REALE, Beitr. z. Klin. d. Tuberk. **82**:180, 1933.

Changes in the leukocytic blood picture in tuberculosis are usually most striking in acute exudative exacerbations. They do not, however, parallel either the anatomic extent of the process or its immunobiologic type. In general, the variations in the total white cell picture are a more delicate index of the evolution of the disease than are the alterations in the polymorphonuclear cells.

AARON EDWIN MARGULIS.

METABOLISM OF WATER AND CHLORIDES IN TUBERCULOSIS. WILHELM GRÜNEWALD, Beitr. z. Klin. d. Tuberk. **82**:189, 1933.

These investigations were stimulated by the lack of a theoretical basis for the salt-free diets of Gerson and of Sauerbruch and Hermannsdorfer. Grünwald determined the water and chloride content of the lymph nodes, kidneys, suprarenal glands, pancreas, spleen, liver, lungs, heart muscle, bones, skin and striated muscles for normal and tuberculous persons. In general, the organs of the latter contained somewhat more water and definitely less chlorides than the normal controls. It is pointed out that such changes are characteristic of all wasting diseases. The conclusion is therefore reached that salt-free diets are not therapeutically rational in tuberculosis.

AARON EDWIN MARGULIS.

METABOLISM OF TUBERCLE BACILLI. L. M. MODEL, J. P. GURJEFF and A. M. PIROGOFF, Beitr. z. Klin. d. Tuberk. **82**:474, 1933.

The authors studied the biochemical changes occurring in a synthetic medium during the growth and autolysis of tubercle bacilli. The following determinations were made: 1. Glycerin and dextrose are energetically destroyed. 2. No protein-split products are formed until, following exhaustion of all assimilable nitrogen, the growth phase ceases and autolysis sets in. 3. The  $p_H$  of the medium decreases during the growth phase but with increasing age of the culture and increasing autolysis rises again, yielding a very typical curve which may be easily differentiated from that given by cultures of paratubercle bacilli, which are invariably alkaline by the end of the six weeks—a difference which may be used diagnostically. 4. Tubercle bacilli can grow under anaerobic conditions. 5. Tuberculin is probably an endotoxin and decreases surface tension.

AARON EDWIN MARGULIS.

SCARLET FEVER WITH PRIMARY INVOLVEMENT OF THE LUNG. HELENE SAWRIMOWITSCH, Beitr. z. path. Anat. u. z. allg. Path. **91**:225, 1933.

Although the earliest localization of the disease process in scarlet fever is usually in the pharynx and tonsils, cases in which the primary lesion is in the skin are not unknown, and a few have been reported in which it was in the larynx or the trachea. The author presents three cases in which the diagnosis of scarlatina could not be questioned and in which there was no involvement of the upper air passages except hyperemia of the pharynx. The primary localization was in the lungs, resulting in a necrotizing lobular pneumonia, in which there were many streptococci and relatively few leukocytes. Each lobular area was surrounded by a zone of fibrinous pneumonia and edema. The character of the inflammatory process is held to be identical with that which occurs in the tonsils in the usual case of scarlet fever. Death occurred early, on the second, fifth and seventh days.

O. T. SCHULTZ.

HISTOLOGY OF TUBERCULOSIS OF BONE. A. N. TSCHISTOWITSCH and S. WINOGRADOW, Beitr. z. path. Anat. u. z. allg. Path. **91**:236, 1933.

In twelve cases of miliary tuberculosis of children and adults the marrow of the diaphysis and epiphyses of the femur and that of the sternum were examined

histologically for the purpose of determining the character of the earliest lesion in tuberculosis of bone. The earliest lesion was a miliary granuloma with a prominent fibrillated reticulum. Necrosis occurred first at the center of the granuloma, and not in the otherwise unchanged marrow as has been asserted by some. Destruction of osseous trabeculae was secondary to necrosis of the granulomas.

O. T. SCHULTZ.

Etiology of Appendicitis. M. GUNDEL, W. PAGEL and F. SÜSSBRICH, Beitr. z. path. Anat. u. z. allg. Path. **91**:399, 1933.

This thorough study of the bacterial etiology of appendicitis is based on thirty-one cases of acute appendicitis, thirty of acute appendicitis with perforation and peritonitis, seventeen of appendical abscess, fifty of chronic appendicitis and seventeen appendixes of the kind that are referred to in the German literature as "stolen appendixes." The work consisted in a bacterioscopic and cultural examination of material from three places in the lumen of the appendix and of material from the throat, a histologic examination of the appendix, with especial reference to the localization of bacteria, a cultural study of the blood just before or after operation, and a bacteriologic study of the stool. The authors conclude that acute appendicitis is an infection by autogenous enterogenous bacteria. The most common causative agent is the enterococcus or the closely related anhemolytic streptococcus. There was no constant or even suggestive relationship between the flora of the appendix and that of the throat. In three cases, however, the same type of pneumococcus was isolated from the appendix as from the throat. A bacterial relationship between acute appendicitis and acute angina is denied. Although the enterococcus is the most important cause of acute appendicitis, in appendical abscess and in peritonitis following perforation the colon bacillus is the most important organism. In these two complications the gas bacillus was also frequently encountered but is believed to have had little part in the inflammatory process. Blood cultures were negative, with the exception of one case in which the colon bacillus was isolated.

O. T. SCHULTZ.

Eosinophil Reaction in Active Tuberculosis. F. A. MICHAJLOW, Virchows Arch. f. path. Anat. **289**:315, 1933.

A decrease in the number of eosinophilic leukocytes in the peripheral blood after the injection of a minute quantity of tuberculin is of diagnostic value in determining the activity or the quiescence of tuberculosis in adults. The quantity of tuberculin must be so small that it will not cause a nonspecific reaction. The eosinophilic cells of blood diluted with Dunger's eosin-acetone mixture are counted in the hemacytometer. Then 0.1 cc. of a 1:100,000,000 dilution of Denys' tuberculin or of a like dilution of Koch's old tuberculin is injected beneath the skin of the shoulder. Thirty minutes after the injection the eosinophilic leukocytes are counted again. A decrease of 5 per cent is considered to show a positive reaction. The reaction is negative, i. e., the number of eosinophils is increased or remains unchanged in persons with inactive tuberculosis, in healthy persons and in those ill of diseases other than tuberculosis. The reaction is positive in adult persons with active tuberculosis and in tuberculous children, and has diagnostic value in differentiating between active tuberculosis and other infections. The reaction is of no value in cases of advanced tuberculosis, in those in which tuberculin is being or has been administered therapeutically, and in those in which the initial eosinophil count is low. The reaction is ascribed to the protein fraction of the tuberculin. In echinococcus disease the injection of minute quantities of echinococcus liquid yields a similar diagnostic reaction.

O. T. SCHULTZ.

Reactive Reticulo-Endotheliosis. V. UHER, Virchows Arch. f. path. Anat. **289**:504, 1933.

A child, aged 21 months, died of streptococcal sepsis seventeen days after the onset of the illness with acute angina. Fourteen days before death the leukocyte

count was 11,000, with 22 per cent lymphocytes and 3 per cent monocytes. The reticulo-endothelial system of the spleen, lungs, lymph nodes and intestine at necropsy was in a state of reactive hyperplasia. The liver and bone marrow took very little part in the reactive process.

O. T. SCHULTZ.

**EXTRAPHARYNGEAL SCARLET FEVER.** A. M. TROIZKAJA-ANDREEWA, *Virchows Arch. f. path. Anat.* **289**:718, 1933.

Cases of scarlet fever in which the primary localization is in a part of the body other than the pharynx and tonsils have been long known and frequently described as surgical, puerperal and wound scarlet fever. According to the author, histologic study of the primary focus is wanting in most such cases. He therefore presents a detailed histologic study of the local lesion in six of sixteen patients with extrapharyngeal scarlet fever who died in a hospital for infectious diseases of children in Leningrad. In eight of the patients a clinically typical attack of scarlet fever followed burns of the skin. In the remainder the point of entrance was a cutaneous eruption or a slight injury of the skin that became infected. The local lesion was characterized by widespread necrosis and purulent infiltration of the skin and underlying fat. Streptococci were numerous in the inflamed tissues. The regional lymph nodes revealed necrosis and the presence of fibrinous exudate in the sinuses. That extrapharyngeal scarlet fever is true scarlet fever must be determined by clinical and epidemiologic considerations. It differs from true scarlatina in no way except that the period of incubation may be shorter and the mortality higher. That a case of scarlet fever is extrapharyngeal in origin must be determined by the absence of acute inflammatory reaction in the tonsils and cervical lymph nodes and by the presence of a necrotizing, purulent process elsewhere.

O. T. SCHULTZ.

**INFLUENCE OF SOME END-PRODUCTS OF METABOLISM ON EXPERIMENTAL TUBERCULOSIS OF GUINEA-PIGS.** PIETRO RONDONI, *Ztschr. f. Immunitätsforsch. u. exper. Therap.* **77**:264, 1932.

Young animals (from 200 to 300 Gm.) were infected with tubercle bacilli of rather low virulence and given repeated injections of small doses of uric acid, creatine or histamine. Uric acid had an aggravating influence and creatine a slightly inhibitory effect on the tuberculous infection. The infection was not influenced by the injections of histamine, but, on the other hand, susceptibility to histamine seemed elevated in the tuberculous animals.

I. DAVIDSOHN.

### Immunology

**EXPERIMENTAL RESEARCH ON YELLOW FEVER.** J. LAIGRET, *Arch. Inst. Pasteur de Tunis* **21**:412, 1933.

In a second series of seven persons vaccinated with the mouse virus of yellow fever, fever developed in three. In one only a febrile reaction occurred; the other two presented various symptoms. Although efforts to recover the virus were unsuccessful, it was believed that the mouse virus, which was not fixed, was not sufficiently attenuated to induce a desired completely silent infection. Attenuation similar to that secured by drying the virus of rabies was accomplished by holding the virus at between 10 and 20 C. Two preparations, one a suspension of glycerin held at ordinary temperature and the other a dried material, failed to kill mice, but induced the formation of protective substances. They had not been tried on man.

FROM THE AUTHOR'S CONCLUSIONS.

**HEMOLYSIS: I-V,** HECTOR DIACONO, *Arch. Inst. Pasteur de Tunis* **21**:557, 579 and 594, 1933; **22**:47 and 212, 1933.

In a series of articles a complete review of the subject of hemolysis is presented, including original research. The first article deals with hemolysis resulting

from distilled water and from other physical factors. The second is concerned with the action of organic and inorganic chemical agents of many types. In the third various bacterial agents are considered, as well as toxins and venoms of vegetable and animal origin. The following study takes up all phases of hemolysis resulting from the use of lytic antibodies and complement. In a fifth article original studies are presented, dealing chiefly with the various factors, physical, chemical, metabolic, etc., affecting the production of antibodies and the mechanism of reaction. The series is concisely presented, although it forms the equivalent of a monograph of 255 pages.

M. S. MARSHALL.

**BLOOD GROUP FERMENT AND ELIMINATION OF BLOOD GROUP SUBSTANCE.**  
E. WITEBSKY and T. SATOH, *Klin. Wchnschr.* **12**:948, 1933.

The stools of adults do not show any group-specific reactions. However, meconium and the stools of the new-born are rich in group A, which disappears during the course of the first year. Schiff believes that this deficiency of group characteristics in the adult is due to the presence of a special blood group ferment. Witebsky and Satoh obtained a Berkefeld filtrate of stool extract and demonstrated that it has a destructive action on group A in saliva. This blood group ferment develops in the stool during the course of the first year.

JACOB KLEIN.

**THE DIAGNOSIS OF LEPROSY BY THE REACTION OF WITEBSKY.** YOSHIO AOKI  
and KITOSHI MURAO, *Ztschr. f. Immunitätsforsch. u. exper. Therap.* **79**:365,  
1933.

Witebsky, Klingenstein and Kuhn prepared a highly sensitive antigen from tubercle bacilli. It reacts with serums of tuberculous patients and, according to Brants, also with serums of lepers. Aoki and Murao checked the results of Brants on serums of thirty lepers and found the original antigen of Witebsky highly sensitive. They also prepared an antigen from lepromas according to the directions given by Witebsky for the preparation of the antigen from tubercle bacilli. The antigen from leproma was less sensitive and less specific than that from tubercle bacilli.

I. DAVIDSOHN.

**CHEMICAL NATURE OF ANTIGENS EMPLOYED IN THE DIAGNOSIS OF SYPHILIS.**  
E. BALBI, *Ztschr. f. Immunitätsforsch. u. exper. Therap.* **79**:372, 1933.

Ö. Fischer showed that when an alcoholic extract of beef heart is treated with aluminum hydroxide it loses its ability to fix complement in the presence of syphilitic serums. The reacting substance could be recovered from the adsorbent by washing it with benzene, and the latter reacted as well as the original extract. Balbi confirmed the results of other authors concerning the antigenic qualities of alcoholic extracts of beef heart when injected into rabbits and the failure of extracts which were adsorbed with aluminum hydroxide to produce lipoid antibodies in the animal. In addition, Balbi reports the production of complement-fixing antibodies following injection into rabbits of benzene washings of aluminum hydroxide, with which an alcoholic extract of beef heart was adsorbed. The immune serum reacted with the native extract and with the benzene washings but not with the adsorbed extract. The results indicate a far reaching parallelism of reactions in vivo and in vitro.

I. DAVIDSOHN.

**THE M AND N COMPONENTS.** GUENTER BLAUROCK, *Ztschr. f. Immunitätsforsch u. exper. Therap.* **79**:377, 1933.

The article begins with a review of publications on the inheritance of the qualities M and N. Seven hundred and fifty-four families with 2,179 children have been studied (559 families and 1,251 children in Europe and the rest in the United States). All the reports, including Blaurock's study of 80 families, confirm

the original assumption of Landsteiner and Levine that the qualities M and N are inherited as a simple mendelian pair of two allelomorphic genotypes. The few (8) exceptions can well be attributed to illegitimacy. Blaurock advocates the recognition of these two qualities for medicolegal purposes. He recommends the absorption of anti-N immune serums for purposes of purification at room temperature in addition to the absorption in the incubator. A technic of agglutination on thick concave slides is described.

I. DAVIDSOHN.

THE CHEMICAL NATURE OF ANTIGENS EMPLOYED IN THE DIAGNOSIS OF SYPHILIS: III. Ö. FISCHER, Ztschr. f. Immunitätsforsch. u. exper. Therap. 79:391, 1933.

Fischer reports additional studies on the nature of the chemical changes occurring when the reacting substance of the alcoholic extract of beef heart is removed by means of aluminum hydroxide and when it is then again liberated by washing the aluminum in benzene. The washings contain more fatty acids and particularly more reducing substances and less phosphatide and nitrogen than the original extract.

I. DAVIDSOHN.

THE MECHANISM OF THE URTICARIAL IDIOSYNCRASY TO EGG WHITE IN THE ECZEMATOUS CHILD. WERNER JADASSOHN and FRITZ SCHAAF, Ztschr. f. Immunitätsforsch. u. exper. Therap. 79:407, 1933.

Dialysates of egg white are known to be urticariogenic in the allergic eczematous child. Opinions differ as to whether the reacting substance is of protein nature, or, as Jadassohn and Schaaf and others maintain, a nonprotein substance. When the proper technic is employed, no protein substances pass into the dialysate. An analysis of the reports to the contrary discloses faults in the technic of dialyzing or in the methods of determining the presence of protein substances in the dialysates. The comparison, by means of the Prausnitz-Küstner technic, of the urticariogenic potency of egg white and of dialysate revealed that the latter reacted in quantities which could not contain the minimum concentration of protein found necessary in quantitative tests with egg white. By means of the Prausnitz-Küstner technic it was possible to demonstrate that chicken egg white contains at least two antigenic substances; one of them is also present in duck egg white. Both pass through the dialyzing membrane. The blood serum of eczematous children with urticarial hypersensitivity to egg white contains substances which can neutralize the urticariogenic properties of egg white. By means of the Schultz-Dale technic trichophytin produced anaphylactic phenomena in dilutions which made it probable that that reaction was not due to protein substances. Jadassohn and Schaaf conclude that urticarial hypersensitivity and anaphylaxis are closely related.

I. DAVIDSOHN.

THE DIAGNOSIS OF WEIL'S DISEASE WITH COMPLEMENT FIXATION AND PRECIPITATION. W. GAEHTGENS, Ztschr. f. Immunitätsforsch. u. exper. Therap. 79:428, 1933.

The antigen was prepared from cultures of Spirochaeta icterogenes on Uhlenhuth's or Korthof's mediums. Cultures of proper density were washed and suspended in phenolized physiologic solution of sodium chloride. The complement-fixation test gave results comparable with those of the older agglutination test, but only in about three to four times stronger dilutions of the serum. The methods complement each other. Precipitation tests based on the use of the benzochol extract of Sachs and his associates and on the clearing reaction of Meinicke gave results similar to those of the two previously described procedures but were unfit for differentiation of Weil's disease from syphilis.

I. DAVIDSOHN.

THE ANAPHYLACTIC SHOCK AS "MODEL" EXPERIMENT FOR ALLERGIC DISEASES.  
P. MANTEUFEL and R. PREUNER, Ztschr. f. Immunitätsforsch. u. exper. Therap. 80:65, 1933.

The use of animals for the study of the therapy and prophylaxis of human allergy lacked comparative value when intravenous or intracardiac injections were necessary for the production of the anaphylactic shock. On the other hand, the Arthus phenomenon was not sufficiently sensitive. Manteufel and Preuner confirmed the results of previous authors who introduced the inhalation experiment. It permits the reproduction of anaphylactic reactions which resemble the disease in man both in the circumstances under which it occurs and in the symptoms. There is no need for repeated experiments to determine the active dose, and the same animal can be employed repeatedly.

I. DAVIDSOHN.

INFLUENCE OF CONCENTRATION ON COMPLEMENT-FIXATION. EDITH SUSSMANOWITZ, Ztschr. f. Immunitätsforsch. u. exper. Therap. 80:95, 1933.

The fixation of complement by protein antiserums was directly proportionate to the concentration of the complement and inversely proportionate to the volume of the complement dilution, (the volume of antigen and of antiserums remaining unchanged). That effect was evident only during the phase of complement fixation (primary incubation). Subsequent addition of physiologic solution of sodium chloride did not affect the intensity of the complement fixation. The effect of the concentration was particularly noticeable when secondary incubation was prolonged, indicating that also the stability of the complement was increased in higher concentrations. When the period of the primary incubation was shortened, the difference was marked also after a short period of secondary incubation. The specificity of the reaction was not affected by the concentration of the complement. The intensity of the complement fixation was further increased by lowering the concentration of the sodium chloride during the period of primary incubation and by decreasing the quantities of the antiserum. A similar but much less marked increase in the intensity of the reaction was observed with antibacterial immune serum, but when lipoid antiserums were employed, the concentration of the complement and the total volume did not affect the intensity of the complement fixation.

I. DAVIDSOHN.

THE SEROLOGIC SPECIFICITY OF SALIVA. ERNST WITEBSKY and WERNER HENLE, Ztschr. f. Immunitätsforsch. u. exper. Therap. 80:108, 1933.

Immune serums produced in rabbits treated with centrifugated and inactivated saliva reacted specifically with their homologous antigen in the complement-fixation test. There was some rather irregular reaction also with human serum. Some antiserums produced by injection of human serum did not react with saliva in the complement-fixation test. The specific salivary substance was highly thermo-resistant. The enzyme which destroys the group-specific substance of the saliva acts similarly though not quite as regularly on the salivary specific substance.

I. DAVIDSOHN.

GROUP-SPECIFIC DIFFERENTIATION OF THE BLOOD OF HORSES. S. SCHERMER and A. KAEMPFER, Ztschr. f. Immunitätsforsch. u. exper. Therap. 80:146, 1933.

By proper experiments on cross-absorption, six group-specific qualities in the red blood cells and six corresponding iso-agglutinins in the serum were established. The A  $\alpha$  shows a differentiation similar to that existing in man: a highly sensitive A ( $A_1$ ) and a weakly sensitive  $A_2$ ; a strong  $\alpha$  and a weak  $\alpha$ . The  $\alpha_1$  may coexist with  $A_2$  in the same horse without an interaction. Such a combination is physiologic. There is no qualitative difference between the two A qualities. The four new pairs of agglutinable and agglutinating qualities are X-x, Y-y, Z-z and V-v. The V-v pair is characterized by a very low titer. The finding of the new

qualities makes it possible to classify the blood of horses without the acceptance of hypothetic subgroups. Schermer and Kaempffer emphasize that they employ the terms A and B as indicating in the horse properties similar to but not necessarily identical with the corresponding human qualities.

I. DAVIDSOHN.

**THE ELIMINATION OF GROUP AND SALIVA SPECIFIC SUBSTANCES.** WERNER HENLE, *Ztschr. f. Immunitätsforsch. u. exper. Therap.* **80**:171, 1933.

Henle confirms the observation of Schiff and Sasaki that human beings can be divided into two groups: those who eliminate their group-specific qualities in the saliva, and those who do not. There is no relation between the ability to eliminate these substances and the presence in the saliva of ferment which are known to destroy the blood group qualities. The specific antigenic salivary substance and the species-specific quality are found in the saliva of the eliminators of the group-specific qualities as well as in that of the noneliminators. The existence of a specific quality "O" in the saliva, as maintained by Schiff, is questioned. It may be mimicked by a demonstrated nonspecific action of concentrated saliva.

I. DAVIDSOHN.

**BLOOD GROUPS IN THE INSANE.** I. SOMOGYI and L. ANGYAL, *Orvosi hetil.* **76**:265, 1932.

A comparison of the percentage distribution of the blood groups of 608 male and 392 female patients with psychoses with the results of the investigations of Verzar and Weszeczky on the distribution of these groups among healthy Hungarians gave the following results: 1. There are no essential differences between the distributions of the blood groups in the healthy and the insane population. 2. There is no characteristic distribution for the different types of psychoses. 3. Persons with nervous and psychotic diseases of syphilitic origin show the same distribution of blood groups as the healthy population. 4. Identity or differences of blood groups do not influence the time of incubation or the type of fever in patients inoculated with malaria. 5. Malarial treatment does not result in any alteration of the groups. 6. No correlation is found between psychopathic heredity and the transmission of blood groups.

WILLIAM FREEMAN.

### Tumors

**A NEW TRANSPLANTABLE TUMOR OF THE RAT.** D. P. SECOF, *Am. J. Cancer* **19**:1, 1933.

The tumor appeared in the supra-orbital region. It has been transplanted through nineteen generations, about 30 per cent of the transplants taking. It grows best in the peritoneal cavity. In structure the tumor resembles a carcinoma, but the type cell suggests that it may belong in the endothelioma or myeloma group. No differentiation in structure has taken place during transplantation. This paper records data on transmission and structure.

**EXTRAGENITAL CHORIONCARCINOMA IN THE MALE.** H. G. HEANEY, *Am. J. Cancer* **19**:22, 1933.

A review of the literature disclosed reports of 131 cases of chorioncarcroma in males. In more than 90 per cent the growth arose primarily in the testicle. In some cases the origin was doubtful, but in at least 7 it seems to have been definitely extragenital and to this group Heaney adds an eighth case. This was a tumor of choriocarcinomatous structure arising in the retroperitoneal tissue of a man 40 years of age. Of the numerous theories of the histogenesis, that assigning the origin to the urogenital anlage appears to be the most satisfactory explanation in this case.

FROM THE AUTHOR'S SUMMARY.

MULTIPLE HEMANGIOMAS IN AN INFANT. A. C. TAYLOR and E. MOORE, Am. J. Cancer 19:31, 1933.

In an infant multiple hemangioma involved the skin, liver and lungs. The question of metastasis is discussed, but no definite conclusions are reached.

THE ABDERHALDEN REACTION IN THE DIAGNOSIS OF MALIGNANT TUMORS. M. SANCHEZ, Am. J. Cancer 19:40, 1933.

Employing nine substrata, Sanchez made 228 Abderhalden tests, with definite results in 185. The most clearcut reactions were obtained with substrata prepared from a carcinoma of the stomach and a sarcoma of the uterus. With a mixture of these two tumors (substratum D) 152 definite reactions were obtained in 127 tests, with a percentage of error of 8.6. Including the uncertain readings and the tests without result, the percentage of error was 23.7. The failures include those due to defects of apparatus as well as those due to the character of the material tested.

FROM THE AUTHOR'S SUMMARY.

KERATINIZING EMBRYONAL NEPHROMA OF THE KIDNEYS OF THE CHICKEN. W. F. FELDMAN and C. OLSON JR., Am. J. Cancer 19:47, 1933.

Bilateral embryonal renal tumors were observed in a cross-bred cockerel, 10 months of age. The right kidney was practically obliterated by a tumor that weighed approximately 100 Gm. The tumor of the left kidney was much smaller and was confined to the region just under the ureter. Metastasis had not occurred. Microscopically these tumors disclosed a variety of cellular elements characteristic of a nephrogenic form of neoplasm of the fetal type. Adenomatous structures were common, and the cells of many of these were undergoing keratinization, with the formation of numerous cornified nodules or epithelial pearls. In the larger tumor cornification was the most prominent feature.

AUTHORS' SUMMARY.

CARCINOMA OF THE SKIN IN CHILDHOOD. JAMES R. LISA, Am. J. Dis. Child. 46:561, 1933.

A boy, 10 years of age, of Polish parentage, had a pedunculated mass on the left side of the lower lip occurring below the mucocutaneous border. It had first been noted about six months previously as a pinhead-sized, cherry-red, hard growth, and it had gradually grown to the size of a pea. Microscopically the tumor was covered by thin squamous epithelium. The rete pegs displayed a marked tendency toward downward growth and branching. At the tips of the pegs the epithelial cells were large and irregular in size and shape; they displayed hyperchromatic nuclei, and the basement membrane was absent. Between the pegs the epithelium was normal. Throughout the dense fibrous subcutaneous tumor mass were scattered, individually and in nests, cells of the same character as those at the tips of the pegs, showing an extreme degree of anaplasia. Within many of the lymphatic channels were masses of very large anaplastic cells. Mitoses were infrequent. The patient has apparently remained free from recurrence or metastasis for one year but is still under observation.

RALPH FULLER.

GRANULOMA CELL HYPERPLASIA OF THE OVARY. J. I. BREWER and A. O. JONES, Am. J. Obst. & Gynec. 25:505, 1933.

Three cases of ovarian tumor due to granulosa cell hyperplasia are described. In all three the growth was associated with uterine bleeding, although two of the cases occurred after the menopause. Metastases were not found, nor have recurrences been observed. It is concluded that growth of this type is not a tumor, but rather hyperplasia of the granulosa cells, which have their origin in embryologic rests of the germinal epithelium. The growth is probably benign, and produces a hormone that is the cause of the uterine bleeding.

JACOB KLEIN.

THE INFECTIVE AGENT IN TUMOUR FILTRATES: A FURTHER INVESTIGATION BY MEANS OF ANTISERA TO NORMAL TISSUES. W. E. GYE and W. J. PURDY, Brit. J. Exper. Path. **14**:250, 1933.

In 1931 we published evidence that when a Fujinami tumor passes from fowl to duck something essential to infectivity changes, since filtrates can no longer be neutralized by immune serum prepared in goats by means of minced fowl embryo. In the first part of the present paper the reverse process is directly demonstrated by means of serum taken from a goat immunized with minced duck embryo. In the later parts of the present paper it is shown that serum taken from a goat immunized by means of fowl red cells will neutralize extract of fowl-grown Fujinami tumor. It is shown also that this same immune serum does not affect the potency of extract of duck-grown Fujinami tumor. Serum prepared in a goat by means of fowl plasma is shown not to affect the potency of filtrate prepared from Fujinami tumor, whether fowl or duck; and the same is shown to be true of serum prepared by means of duck plasma. **AUTHORS' SUMMARY.**

THE HETERO-TRANSFER OF TWO FILTERABLE TUMOURS: AN INVESTIGATION BY MEANS OF IMMUNE SERA. W. J. PURDY, Brit. J. Exper. Path. **14**:260, 1933.

A serologic method dependent on the published observations of Gye and Purdy has been used in a reinvestigation of the way in which daughter-tumors are formed when tissue from fowl-grown Rous and Fujinami tumors is injected into ducks. The new method shows that in adult ducks new Fujinami tumors arise solely because cells of the host become infected and multiply. It shows also that in ducklings Rous sarcoma 1 is propagable solely because the tumor cells of the inoculum become established and multiply; no infection of host cells takes place. Thus by an independent method conclusions drawn from observations made on ducklings which had received an injection of embryo tissue have been confirmed. Gye and Purdy (1931) concluded that the species-specific element which is present in an active tumor filtrate and is necessary for infectivity must be derived from the tumor cells themselves, and not from normal tissues of the host. The accuracy of this conclusion is proved by new facts. **AUTHOR'S SUMMARY.**

ACTIVE IMMUNIZATION OF PHEASANTS AGAINST FOWL TUMOURS. C. H. ANDREWES, J. Path. & Bact. **37**:17, 1933.

Rous sarcoma 1 has been propagated in series through four pheasants, and transplantation could probably have been carried on indefinitely. In addition to the tumors mentioned in an earlier paper, two other fowl tumors (Begg's MH 2 and Baker's BS 1) have been transplanted into pheasants. Pheasants resisting repeated inoculations of tumors to which they are relatively refractory are subsequently found to be immune to filtrates or cells of Rous sarcoma and Fujinami sarcoma. Evidence is given that this immunity depends primarily on immunity to tumor virus antigen; immunity to fowl protein may, however, play a subsidiary rôle. **AUTHOR'S SUMMARY.**

FURTHER SEROLOGICAL STUDIES ON FOWL-TUMOUR VIRUSES. C. H. ANDREWES, J. Path. & Bact. **37**:27, 1933.

Serums from fowls bearing the fibrosarcomas MH 1 and CT 10 neutralize filtrates of the following tumors: fibrosarcomas MH 1 and RF 11, spindle cell sarcomas RF 4, BS 1 and Rous 1, and endothelioma MH 2. They fail to neutralize the virus of Fujinami's myxosarcoma. On the other hand, ducks recovered from Fujinami tumors and hyperimmunized have serum which will neutralize not only Fujinami virus but also, though less readily, Rous, MH 2, BS 1 and RF 4 viruses. Rous and Fujinami viruses can be shown to be antigenically different from one another. Of the tumor viruses studied, Fujinami's is neutralized only by homo-

logous (anti-Fujinami) serum; Rous 1 is readily inactivated by many heterologous serums; RF 4, RF 11, MH 2 and BS 1 stand in an intermediate position. The few "normal" fowl serums with neutralizing properties have proved active particularly against Rous virus, less so against RF 4, RF 11, MH 2 and BS 1, and not at all against Fujinami virus. The fowl tumor viruses which have been thoroughly studied all have some degree of antigenic relationship, but no two have yet been found to be serologically identical. They are probably interrelated much as are the members of some groups of bacteria.

## AUTHOR'S SUMMARY.

VITAL STAINING OF THE ROUS SARCOMA. A. HADDOW, J. Path. & Bact. 37:149, 1933.

By the use of the intravital staining technic it is shown that the tumor appearing after an inoculation of active cell-free filtrates of sarcoma is derived from previously normal cells under the influence of the agent. From the morphology of the cell and its content and distribution of segregated dye it is shown that the unit initially affected is the free histiocyte, and that this must be regarded as the parent cell of the Rous sarcoma.

## AUTHOR'S SUMMARY.

HISTOLOGIC STUDIES OF CARCINOMA OF THE CERVIX AFTER TREATMENT WITH RADIUM. GOFFREDO FROLA, Arch. internat. de méd. expér. 8:289, 1933.

Twenty-four hours after the beginning of the treatment with radium typical mitosis ceased; atypical mitosis was considerably diminished also. Pyknotic changes increased markedly for several days. At the end of the treatment there was a modification in the volume, form and color of the nucleolus. The size of the cells gradually increased after treatment, slightly in adenocarcinoma, but considerably in epidermoid carcinoma. The cytoplasm showed cloudy swelling and vacuoles; the nucleus was distorted by pyknosis, swelling, vacuolation or lysis. The connective tissue became more dense, and the inflammatory reaction, which at first may have been predominantly lymphocytic, became plasmocytic and finally leukocytic. The number of eosinophils increased toward the end of the treatment; this may have been due to irritation by lactic acid and was of no particular significance. The blood vessels were practically always affected by radium; there was proliferation of the intima as well as thrombosis; frequently there was fibrinoid degeneration of the media. Resorption of the necrotic tumor occurred by lysis and by the action of microphages and macrophages. This was followed by sclerosis of the connective tissue. Giant cells similar to those around foreign bodies were also seen. The radium caused a hyperplasia of the mucous glands of the cervix. These glands are resistant to radium and frequently undergo anaplasia or metaplasia. Even after preliminary treatment with x-rays, radium does not destroy mitosis (typical and atypical).

JACOB KLEIN.

THE PATHOGENESIS OF MYXOMA. T. GRECO, Tumori 7:134, 1933.

Pure myxoma originates from embryonic tissue. In the myxosarcoma group only the minority can be considered pure malignant myxoma, the majority being sarcoma with a few islands of degenerated mucoid tissue. E. VON HAAM.

CARCINOSARCOMA OF THE PROSTATE GLAND. C. PANA, Tumori 7:244, 1933.

A man 59 years old had a tumor of the prostate gland showing the histologic picture of small round cell sarcoma and adenocarcinoma. A metastasis in the lymph glands showed the structure of round cell sarcoma. E. VON HAAM.

CYTOLOGIC STUDIES OF MALIGNANT TUMORS. U. C. BAGOZZI, Tumori 7:266, 1933.

Golgi's apparatus and the mitochondria were studied in tumor cells of man and animals. It was found that the Golgi apparatus in cells of benign tumors

and tumors of low grade malignancy is similar to that in normal cells, and that it is only in the cells of very atypical and very malignant tumors that the structure is different. The mitochondria were found to be of a typical cytoplasmic formation, and no difference between the structure of normal cells and that of malignant cells could be detected.

E. VON HAAM.

IMPORTANCE OF THE MALIGNANCY INDEX OF HUEPER AND SCHMITZ IN THE TREATMENT OF CARCINOMA OF THE UTERUS. M. STRANI, *Tumori* 7:289, 1933.

The following characteristic signs mentioned in the malignancy index of Hueper and Schmitz are of great significance in the prognosis and treatment of carcinoma of the uterus: (1) the type of carcinoma and the degree of maturity or differentiation; (2) the type of the infiltrating growth; (3) the size and the shape of the single cells; (4) the sharp demarcation of the limits of the single cells; (5) the irregularity of the shape and size of the cell nucleus, and (6) the number and especially the atypical appearance of the cell mitoses.

The relationship between the nuclear substance and the plasma, the vascularization of the tumor and the degree of inflammatory reaction are of little value for prognosis and therapy.

E. VON HAAM.

CYLINDROMATOUS TUMOR OF THE PAROTID GLAND. AARNO SNELLMAN, Arb. a. d. path. Inst. d. Univ. Helsingfors 7:11, 1933.

A thorough microscopic study of five cases of so-called parotid cylindroma (adenocarcinoma?) was made with particular attention to the parenchyma and the hyaloid formations in the stroma. The latter structures are thought to be due to a combination of the parenchymal secretions with the stroma, which results in an irreversible reaction.

JACOB KLEIN.

CELL STRUCTURE OF RODENT ULCER. C. THESLEFF, Arb. a. d. path. Inst. d. Univ. Helsingfors 7:51, 1933.

A microscopic study of forty-eight specimens of rodent ulcer was made. Solid basal cell carcinoma occurred in more than half the cases. Hyaline pearls and concentric cell formations were frequent. In seven instances a metatypical structure was observed in which the cells were rich in plasma and contained radial structures. The stroma was vascular in these specimens. The presence of plasma cells, granulocytes, lymphocytes and mast cells indicated secondary infection. With special silver stains a definite network was demonstrated, as well as nuclear formations. There are included numerous illustrations, as well as a review of important theories explaining the pathogenesis of rodent ulcer.

JACOB KLEIN.

SYMMETRICAL SQUAMOUS CELL CARCINOMA IN SCARS OF BURNS ON THE LEGS. GEORG ARNDT, *Beitr. z. klin. Chir.* 157:305, 1933.

Symmetrical bilateral carcinoma of the extremities developing in scars from burns is reported for the first time. The 44 year old woman had large cylindromatous ulcers on both legs, extending from the region of the ankle to the middle of the leg. The burns occurred 41 years previously. Ulceration developed in the scars after she was 26 years of age. The ulcers healed during several pregnancies, but finally increased in size. Both legs were amputated, and the patient was without recurrence one year after operation. Histologically the tumors were ripe squamous cell carcinomas. Ninety-nine instances of cancer in scars from burns have been collected from the literature. The occurrence in men has been three times as frequent as in women. The average age was 47 years. Carcinoma may develop shortly after a burn or after intervals as long as sixty-nine years. Like other

carcinomas of the skin that developing from the scar of a burn is the least malignant form of squamous cell carcinoma and offers a fair prognosis (cures in 62 per cent of the cases).

G. ALEXANDER HELLWIG.

GROWTH OF HUMAN TUMORS IN VITRO. Z. ZAKRZEWSKI and W. KRASZEWSKI, Klin. Wchnschr. **12**:1495, 1933.

Cultures of human tumor tissues were made in Carrel flasks. The solid phase was coagulated chicken plasma washed with Tyrode solution; the liquid phase was diluted human serum with heparin. In cultures of twelve different tumors both tumor and stroma elements grew, the latter cells with greater activity. All of the tumor tissues investigated grew well in cultures for from two to four months. These results oppose the concept of a specific tumor-stimulating substance. As has been stated previously, apart from fundamental contrasts between normal and tumor cells no further qualitative differences can be demonstrated. In spite of unsuccessful inoculation of these tissues in human hosts the tissue cells, in their cultural behavior, are malignant.

E. F. HIRSCH.

ACTIVE COLLOID IN A METASTATIC THYROID CARCINOMA. R. B. ENGELSTAD, Ztschr. f. Krebsforsch. **39**:369, 1933.

A patient, 82 years of age, had had for twenty years a goiter without perceptible evidence of change. There developed a metastatic lesion of the skull cap, histologically evidently a carcinoma of the thyroid gland. Active colloid could be demonstrated in this, in a quantity of over 0.25 mg. per gram.

H. E. EGGERS.

## Society Transactions

PATHOLOGICAL SOCIETY OF EASTERN NEW YORK

ARTHUR W. WRIGHT, *Secretary*

*Regular Meeting, Oct. 13, 1933*

J. SCHLEIFSTEIN, *Presiding*

### GENERALIZED METASTATIC MELANOMA. V. W. BERGSTROM.

A married white woman, aged 49, was first taken ill nine weeks before death. She complained of nausea and vomiting as well as an indefinite feeling of distress without pain. There was one fainting spell with unconsciousness four weeks before death. No paralysis or convulsions were noted, but there was slight spasm of the left eyelid and the pupils reacted slowly to light. Moderate edema of the ankles improved with rest in bed. Nervousness and restlessness were marked. The pulse rate averaged about 36 per minute but occasionally went as high as 100; the temperature never went above 100 F.; the respirations did not exceed 26 per minute.

The hepatic dulness reached to the brim of the pelvis. Innumerable bluish-gray, shotlike nodules developed in the skin and mucous membranes over the entire body less than two weeks before death. The nails and conjunctivae were not involved however. Examination of an excised nodule showed it to be a melanoma. No pigmented warts or moles could be found, although there were a few simple non-inflamed warts on the back. There was likewise no evidence of tumor in the eyes. The urine contained a small amount of albumin and a moderate number of granular and hyaline casts. The Wassermann test was negative. An electrocardiogram indicated extreme myocardial damage.

At autopsy every tissue except the cardiac valves, eyes and nails was found to be involved in metastatic pigmented tumors. Particularly massive nodules were found in the medullae of the suprarenal glands, in the hypophysis apparently involving both lobes and the intermediary portion, and in the posterior portion of the thalamus just beneath the corpus callosum.

This case is reported because of the speed and tremendous extent of the metastases, and because no primary focus could be demonstrated. In view of the extensive involvement of both suprarenal glands and the malignant appearance of the medulla of one of them it is thought that the primary melanoma probably originated in one of these glands.

### A LARGE GALLSTONE EXTRUDED AT THE UMBILICUS. ELLIS KELLERT.

A married woman, 60 years old, entered the clinic and stated that two years previously she began to experience attacks of gastric distress and vomiting. These symptoms continued at irregular intervals, usually weeks apart. The bowels were regular and the feces were normal in color. Recently the periods of distress had been more frequent and were associated with severe pain in the upper right quadrant of the abdomen. She entered the hospital during one of these attacks. There was severe epigastric pain radiating to the right lower quadrant of the abdomen, with a rapid pulse and fever.

The patient appeared slightly jaundiced, and a nonfluctuating mass 12 by 15 cm. in size was found in the right upper quadrant of the abdomen. The leukocyte count was 17,600 per cubic millimeter, and the polymorphonuclear leukocytes were 78 per cent. Other laboratory tests were negative. A roentgenogram disclosed

obstruction in the first portion of the duodenum. After several days the patient felt more comfortable and left the hospital.

On her recent visit to the clinic she directed attention to a small, black, hard area involving the umbilicus. This object proved to be a calculus, which was readily extracted by means of a thumb forceps. After removal of the calculus yellow bile discharged from the opening, and a sinus tract to the region of the gallbladder was probed.

The calculus was somewhat oval in outline and measured 4.5 by 3.5 by 3.5 cm. All surfaces were encrusted with brownish-black, glistening, amorphous material which separated readily on handling. When the calculus was sectioned, the cut surface had a distinctly concentric lamellated character, many yellow and dark brown layers being present. On microscopic examination there was an abundance of cholesterol mingled with bile pigment. No bacteria were found.

#### ENDOSALPINGIOSIS OF THE BLADDER. G. H. KLINCK JR.

A white woman, 40 years of age, complained of irregular attacks of pain in the right lower quadrant of the abdomen, associated with frequency of micturition, burning and pain. She had had menorrhagia and metrorrhagia for two months and had lost some weight. Endometrial curettings showed tuberculous endometritis. Cystoscopy revealed a globular tumor, 3 cm. in diameter, with a wide base, in the right wall of the bladder about 3 cm. to the right of and above the right ureteral orifice. A minute bit of this tumor showed bladder epithelium only, and a report of benign papilloma was made.

Complete hysterectomy was done, at which time it was noted that the right tube was so firmly adherent to the right posterolateral wall of the bladder, just opposite to the tumor, that it was necessary to cut the tube away from the wall of the bladder. Examination showed tuberculous endometritis and myometritis with a few early tubercles in the adhesions between the right tube and the bladder. The wall of the right tube in the region of the adhesions to the bladder contained numerous glands composed of columnar epithelium surrounded by a small amount of loose stroma. No blood was found in these glands, although many were dilated. Convalescence was uneventful.

The patient returned six weeks later, and the growth in the bladder was removed through a suprapubic cystotomy. The surface was congested and soft, with small cystlike spaces just beneath the surface. Section showed a growth radiating from the wall of the bladder. It had a soft, edematous stroma containing many small cystic spaces toward the periphery. Microscopically the tumor was composed of dilated glands lined by flat to columnar epithelium. The glands contained amorphous granular material but no intact red cells. In the peripheral portions of the tumor there was very little stroma. Toward the base the glands became smaller, the epithelium more columnar and surrounded by dense fibrous stroma. Gland structures could be traced from the base through the wall of the bladder and well into the old scar tissue, the site of the previous salpingectomy. The surface of the mass was covered over by a layer of hyperplastic bladder epithelium. The patient made an uneventful recovery.

This case illustrates another complication of tubal adhesions in which the tubal mucosa invades another organ. Sampson described such lesions in the ovary and uterus, and in this instance the urinary bladder was involved. Endometrial tissue is able to invade these organs in much the same way.

#### EFFECT OF CESIUM CHLORIDE OF TRANSPLANTED TUMORS OF MICE. ARTHUR W. WRIGHT and CLARENCE F. GRAHAM.

A full report of these studies appeared in the *American Journal of Pathology* (9:789, 1933).

## NEURO-EPITHELIOMA OF THE RETINA. J. SCHLEIFSTEIN.

In 1891 Flexner described a tumor of the retina of a type which has since been known as neuro-epithelioma. The case which I present is a typical instance of neuro-epithelioma according to the classic description recorded by Flexner. The parents of a girl 6 months old consulted a physician because they had noticed something the matter with her eye for the last two months. A diagnosis of glioma was made and the eyeball enucleated.

The specimen consisted of an eyeball somewhat larger than normal for the age. The cornea appeared dull. Section through the eye showed a soft, grayish-pink tumor almost completely filling the eyeball.

Microscopic examination showed a compact mass containing sheaths of round cells with deeply staining nuclei and very little cytoplasm. Many mitotic figures were present. Very little fibrous stroma could be observed. Numerous foci of hemorrhage and necrosis were seen. Scattered throughout the tumor were many rosette formations, which were demonstrated particularly well with Mallory's phosphotungstic acid-hematoxylin stain.

## ADENOMA OF THE SWEAT GLANDS. J. SCHLEIFSTEIN.

Adenoma of the sweat glands forms an interesting class of tumors, some of which border on malignancy. The case to be reported appears to belong to the latter group.

A young man, 23 years of age, complained of a lump behind the occiput. The attending physician made a clinical diagnosis of epidermal cyst and removed the mass, which proved to be a small tumor the size of a lima bean. It contained numerous papillary projections. Microscopic examination showed an actively growing sweat gland adenoma malignum of low grade.

## CARCINOMA OF THE ISLETS OF LANGERHANS. V. C. JACOBSEN.

A white American pharmacist, 36 years of age, suffered gastro-intestinal upsets between 1927 and 1930. These were characterized mainly by diarrhea. In 1930 a diagnosis of faulty fat metabolism was made, and the patient was given a diet high in carbohydrate and low in fat. The symptoms continued and in 1932 a diagnosis of diabetes mellitus was made on the basis of glycosuria. He was put on a diabetic regimen and shortly afterward suffered insulin shock with the blood sugar below 25 mg. per hundred cubic centimeters. Roentgen examination suggested a tumor of the head of the pancreas. Operation was not advised.

The patient required constantly increasing amounts of sugar in order to ward off insulin shock. By August, 1933, he required a glass of orange juice every three hours day and night. In the latter part of August he could not be aroused three hours after taking nourishment but regained consciousness after 2 glasses of orange juice, which he required thereafter every two hours. Insulin shock developed suddenly, with the blood sugar 39 mg. per hundred cubic centimeters. Roentgen examination verified the suspicion of a large tumor in the region of the head of the pancreas, and an exploratory laparotomy was done.

The tumor was roughly the size of a small grapefruit and inoperable, but a small bit was removed for biopsy. The liver contained several small nodules of firm white neoplastic tissue. One of these was removed. During the operation the patient received 150 mg. of dextrose intravenously. The blood sugar rose to 527 mg. per hundred cubic centimeters and within nine hours dropped to 93 mg. Thirty-two hundred milligram-hours of radium was applied over the epigastrium.

Following operation it was possible to prevent insulin shock by the administration of 50 Gm. of dextrose every four hours day and night. At present the patient is in fairly good health and has no symptoms so long as the dextrose is taken regularly. He has slight secondary anemia.

Pathologic examination of the tissue removed from the region of the main pancreatic tumor showed narrow cords of epithelial cells slightly squamous in type,

infiltrating fat and connective tissues. No mitoses were seen. The nodules in the liver were very cellular epithelial growths, the cells showing much regularity and no mitoses. Often arranged in large alveoli with a few capillaries in and about them, they bore a striking resemblance to pancreatic islets. Bensley stains showed only beta granules present.

NOTE.—This patient died Dec. 24, 1933, from hemorrhage caused by erosion of the second portion of the duodenum by the carcinoma in the head of the pancreas. The body and tail showed parenchymal atrophy with persistence of islets. Metastases were present in the liver only. Much glycogen was found in the kidneys. The hyperinsulinism was under good control, the blood sugar post mortem being 180 mg. per hundred cubic centimeters. The case will be reported in more detail by Dr. Nelson K. Fromm.

#### EXTREME HEMOLYSIS ASSOCIATED WITH MARKED LIVIDITY AND HEMOGLOBINURIA. J. J. CLEMMER.

An unmarried white woman, aged 30, was admitted to the hospital in a conscious but toxic state. About thirty-six hours previously she had had an abortion. Her chief complaints were pain in the lower part of the abdomen, nausea and numbness of the legs. She had a moderate fever, a rapid pulse with low blood pressure, and a rapid respiratory rate. The uterus was enlarged and soft, and the cervix was dilated. A profuse black bloody vaginal discharge was present. The entire body was a diffuse dusky red. Catheterization of the bladder yielded an ounce (30 cc.) of dark red fluid, which microscopically showed only a few crenated erythrocytes. The urine contained large quantities of hemoglobin. Blood withdrawn for typing was markedly hemolyzed. Thirteen hours after admission, following a brief convulsion, the patient expired.

Postmortem examination revealed a generalized infection with Clostridium Welchii, with the genital tract the probable portal of entry.

The uterus was enlarged and soft. The wall of the fundus was lacerated. A small probe could be passed through it at one point. A few bits of placental tissue were present in the cavity. The entire genital tract was acutely inflamed.

Striking evidence of generalized hemolysis was present. The serous linings were stained uniformly pink. The endothelial lining of the heart and large vessels showed similar staining. The kidneys were intensely deep red.

There was no evidence of gas production in the tissues at the time of necropsy. However, a few days later, bubbles of gas were found situated deeply in some of the organs which had been saved as gross specimens.

This case is presented to emphasize the marked hemolytic action exhibited by some strains of Cl. Welchii. This organism has been known to produce a hemolysin, but the biologic activity usually stressed is its production of gas in vivo and in vitro. Clinically, gas production was not demonstrable in this case. However, the marked hemolysis observed both ante mortem and post mortem was sufficient to suggest the diagnosis of Cl. Welchii septicemia.

#### MEASLES ENCEPHALOMYELITIS. R. J. LEBOWICH.

Five days after the appearance of the rash, this nervous complication of measles developed suddenly in a white child, 4 years of age, with convulsions and muscular twitchings limited at first to the left side of the face, later spreading to the neck and right arm. The child became unconscious, failed to answer questions, and was not responsive to external stimuli. The eyes were drawn to the left. No attempt was made to move the left arm or leg. The spinal fluid was normal. The patient became progressively worse with increasing frequency of muscular spasms over the right and then the left side of the body. Death occurred about forty-eight hours after the initial development of the convulsions.

To the naked eye the only changes of note in the brain were pronounced congestion and edema of the leptomeninges. Microscopically, however, these tissues

showed a slight, intermittent perivascular infiltration by lymphocytes, plasma cells and macrophages. Focal histologic changes were also distributed rather uniformly from the frontal cortex into the upper cervical cord. These changes consisted of perivascular and marginal zones of demyelinization and complete destruction of axis-cylinders located almost always about the small veins. They were of the sharply punched-out type and formed wide sleeves about the vessels for long distances, with extensions along their branches.

The most common cell present in the extra-adventitial infiltrates was the lymphocyte. Plasma cells were common. Frequent active or inactive macrophages associated with occasional polymorphonuclears were also observed, especially in the very severe perivascular lesions. Many astrocytes were present either within the extra-adventitial lesions or at the periphery, often flowing out into the more intact nerve tissue. In general, the ganglion cells were remarkably well preserved and complete necrosis was rare. There were no signs of neuronophagia.

It is of interest to note that in the centrum ovale minute irregular masses of lime salts were occasionally observed in the ground substance, especially in areas of incomplete disintegration. No calcification of the arteries of the type now familiar in chronic epidemic encephalitis was seen.

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NEW YORK PATHOLOGICAL SOCIETY AND NEW YORK  
ACADEMY OF MEDICINE, SECTION OF SURGERY

*Joint Meeting, Nov. 23, 1933*

PAUL KLEMPERER, *President*

SYMPOSIUM ON GASTRIC NEOPLASMS

SOME FEATURES OF THE PATHOLOGY OF PRIMARY CANCER OF THE STOMACH.  
ARTHUR PURDY STOUT.

The malignant neoplasms of the stomach include both carcinomas and sarcomas, the latter forming about 3 per cent of the total number. The vast majority of the carcinomas arise from the mucosal lining or gland cells and have a tendency to form glands and to secrete mucus. The growth tends to advance centrifugally from the focus of origin, i. e., into the lumen and into and along the wall of the stomach. But different cancers display these tendencies in differing degrees, and this fact enables one to divide them roughly into two great groups: (a) the *fungating*, with growth chiefly into the lumen, and (b) the *infiltrating*, with growth chiefly along the wall paralleling the lumen or into the wall away from the lumen. These two groups can be subdivided, depending on the presence or the absence of ulceration.

The fungating group includes about one third of the carcinomas and contains most of the better differentiated, less malignant tumors, which can be successfully resected.

The infiltrating group comprises about two thirds of the carcinomas and contains most of the poorly differentiated, very malignant tumors.

The presence of metastases in the regional lymph nodes is a prognostic sign of the gravest significance no matter what the type of tumor, but an enlarged, hard lymph node does not always contain metastases.

Carcinomas develop at the margins of simple gastric ulcers; a cancer may occur in a stomach which has a simple ulcer in a different part of it. An infiltrating ulcerated carcinoma may simulate a simple ulcer.

The duodenum is seldom invaded by a gastric cancer, and when it is, the invasion is of microscopic extent. However, a portion of a fungating tumor may protrude through the pylorus and stick out into the duodenum.

## DISCUSSION

N. C. Foot: As Dr. Stout says, carcinoma of the stomach might be classified on a histologic or histogenetic basis, this being qualified by various subheadings depending on whether mucus is formed, whether fibrous tissue reacts to the presence of the tumor and similar factors. Unless the surgeon knows the characteristics of each type of carcinoma he can gain little knowledge as to prognosis from the histogenetic diagnosis alone. Dr. Stout's simple classification of carcinoma into tumors that grow outward or fungate and those that grow inward or infiltrate gives one an idea of the gross appearance of the type and an inkling as to the prognosis. Coupled with the prognosis, as based on fungating or infiltrating growth, is Dr. Stout's warning as to the importance of microscopic examination of the regional lymph nodes. Only the microscope can indicate whether there is or is not metastasis to the lymph nodes, and the statement that enlarged nodes do not necessarily contain metastases is borne out by my experience. The converse is equally true: small and apparently innocent nodes may contain small metastases that will develop into dangerous carcinomas. This makes microscopic examination of all nodes removed at operation imperative.

The fact that fungating tumors are less malignant than infiltrating ones goes hand in hand with their differing degrees of differentiation and specialization; those that are forming glands and differentiating toward their normal goal do not tend so much to invade, while those that form distorted travesties of glands, or mere plugs or masses, or even mere gangs of discrete cells, are very poorly differentiated and therefore know no laws and proceed to invade foreign territory without providing themselves with passports. The grading of tumors is based on the degrees of differentiation, so that tumors of the lower grades of malignancy fall automatically into Dr. Stout's outgrowing group, while those of the higher grades come into the ingrowing, or infiltrating, group.

I am not much impressed with the importance of grading tumors, for two reasons. The first one is that one tumor may show areas of differing degrees of malignancy, so that several slides from the same growth may be classified in as many grades. Secondly, the criteria for grading are largely matters of personal impression and individual judgment. If the surgeon is familiar with the criteria employed by his favorite pathologist, grading may mean something to him, but if he takes the grades of a man with whom he is not well acquainted he may easily be misled. Few pathologists take the time to count the well and the poorly differentiated cells in a given tumor. They usually grade a tumor by superficial impression, which makes the whole thing a personal matter. Is it not better plainly to state one's opinion as to whether a growth is markedly, moderately or slightly malignant in its microscopic appearance, and let it go at that?

Dr. Stout has come out courageously for the possibility of a cancer developing in a chronic ulcer, and I am very glad that he has done so. Last year I had a patient who had had a chronic ulcer for about six years; when he was subjected to operation, the microscope revealed a tiny carcinoma developing in the margin of the ulcer, which on gross inspection looked like an ordinary chronic ulcer. It would have been missed if radial sections of the crater and lip, like slices of a pie, had not been made. The simultaneous development of an ulcer and a cancer in the same stomach has been observed by many.

The freedom of the duodenum from carcinoma is indeed baffling; the duodenum and the pylorus are so essentially alike in histologic structure that it seems strange that the one should go almost scot-free while the other constitutes the site of predilection for gastric carcinoma—this in spite of the fact that chronic ulcers occur in each. Boyd mentions this immunity in his textbook and cites Deaver's figures, which show that when carcinoma occurs in the duodenum it is usually found in the second portion near the papilla of Vater, while the first and third segments rank next, in that order. Ewing's figures show that duodenal carcinoma forms 4 per cent of all intestinal cancers.

As to gastric sarcoma, occasionally one meets with a leiomyosarcoma of the stomach that simulates carcinoma clinically, but gives practically no symptoms except hematemesis until it is too late to do anything surgically. It usually occupies the fundus, causes no change in the secretions, is not palpable until it has metastasized, and does not show on roentgen examination. It causes widespread metastasis in the liver and elsewhere, does not invade the nerves, and grows much more slowly than does a carcinoma. Hence the clinical history does not feature pain. The patient outlives his predicted span of life on account of the slow progression of the tumor, and one must be very cautious in giving a prognosis as to time in such cases, after operation has revealed extensive involvement of the liver. I have seen two such tumors in the past decade; both ran a slow and comparatively symptomless course; both occurred at the fundus; both were diagnosed as chronic ulcer, and both progressed slowly to an ultimately fatal outcome. The diagnosis of leiomyosarcoma under the microscope is not easy, for there are other sarcomas that may be confused with it; some of these may be so very embryonal that a recognition of the type cell is well nigh impossible, but this is of interest chiefly to the pathologist; the manifest malignancy of the tumor is not difficult to recognize, and this is the all-important fact in the eyes of the physician and the surgeon.

ARTHUR PURDY STOUT: Dr. Foot mentioned the variations in differentiation that many carcinomas of the stomach show. That is certainly true in my experience. They are not all of one type; they vary, many of them in different parts, and when I have found that, I have formulated the conception that a tumor is as malignant as its most malignant-appearing part; usually a tumor which has some part of it poorly differentiated belongs to the more malignant group.

#### ROENTGEN ASPECTS. ROSS GOLDEN.

Malignant disease of the stomach lends itself to detection by roentgen methods by virtue of its three inherent characteristics, namely, its ability to produce tumor masses that project into the lumen and result in "filling defects," its power to infiltrate the walls, change their contour and interfere with their movements, and its tendency to ulcerate. Dr. Stout described the fungating type, in which the formation of tumor masses predominates, and the infiltrating type, in which the formation of tumor masses may be slight or absent. A tumor that does not infiltrate is benign, e. g., mucous polyps, which are recognized by absence of stiffening of the wall and interference with peristalsis. The lantern slides shown illustrate some of the roentgen manifestations of gastric neoplasms, some of the difficulties in their detection and some problems in differential diagnosis.

In one instance, an extensive infiltrating carcinoma failed to keep the stomach from contracting and expelling large masses of barium because the malignant cells infiltrated between and did not destroy the muscle bundles.

An annular ulcerating carcinoma of the prepyloric region was accompanied by a peptic ulcer a little higher up. The malignant growth bulged through the pylorus into one side of the duodenal bulb.

Peptic ulcers in general differ from ulcerating malignant neoplasms by the fact that their craters are usually smaller and relatively deep, while those of carcinomas are larger, shallow and saucer-like. Peptic ulcers occur on or near the lesser curvature, while malignant growths may occur anywhere. Sometimes on careful palpation irregularities around the margin of the malignant crater or a too extensive stiffening of the wall may be demonstrated. Holmes and Hampton (*J. A. M. A.* 99:905, 1932) have found all ulcerating lesions in the prepyloric region to be malignant. A peptic ulcer should be definitely smaller after three weeks of an adequate dietary regimen. Cases illustrating these points and exceptions to the rule are shown: A patient had an ulcer of the lesser curvature in 1924 which disappeared, an ulcer of the duodenum and of the lesser curvature in 1926 which disappeared, another in 1929 which disappeared, and an ulcerating carcinoma in 1933. An ulcer of the lesser curvature which did not reduce in size under treatment had the histologic characteristics of a malignant growth on

one side of the crater and of peptic ulcer on the other. An ulcer immediately adjacent to the pylorus showed no histologic evidence of malignancy. A large, deep ulcer of the lesser curvature, measuring 5 cm., in a man 75 years old, disappeared in two months; the man was alive nine years later.

The diagnostic differentiation of hypertrophy of the pyloric muscle from early annular carcinoma of the prepyloric region in an adult may be impossible roentgenologically and may be difficult at operation (illustrations given). The impression on the proximal end of the duodenal bulb described by Kirklin and Harris (*Am. J. Roentgenol.* **29**:437, 1933) as characteristic of a hypertrophied muscle may be absent, and a similar impression may be produced by the bulging of a carcinoma through the pylorus.

A carcinoma of the cardiac end of the stomach may be extremely difficult to detect. The importance of examination of the patient in the supine position with rotation from side to side was emphasized.

In three instances, a sarcoma of the stomach produced a round, sharply defined filling defect in which was the shadow of a barium-filled crater.

Syphilis of the stomach is a rare condition which cannot be differentiated from carcinoma by roentgen methods. Lymphoblastoma of the stomach may be differentiated only when two levels in the gastro-intestinal tract are involved. Tuberculosis of the stomach has been reported as simulating carcinoma (Renander: *Acta radiol.* **11**:636, 1930).

In conclusion it may be said that the diagnosis of *early* carcinoma of the stomach is extremely difficult and is necessarily attended with certain risks. The roentgenologist will probably make some false positive interpretations. The surgeon will have to explore on relatively meager roentgen evidence of malignancy and will probably do some resections of benign growths because he could not be sure that they were not malignant. However, by intimate correlation of the efforts of the roentgenologist, the surgeon and the pathologist, the goal may be approached, which is the accurate diagnosis of early carcinoma of the stomach.

#### DISCUSSION

W. H. STEWART: Dr. Golden's statement that to date he has not been able to confirm Dr. Kirklin's opinion that a cup-shaped concave base to the duodenal bulb, with or without an apparent widening of the pyloric canal, indicates hypertrophy has been my experience. My last venture in the realms of pyloric hypertrophy proved disastrous, for the patient had a carcinoma. However, it must be admitted that thickening of the pylorus is a true identity and should give distinct roentgen signs. It is up to roentgenologists to recognize them. The difficulty will be to differentiate hypertrophy from malignancy.

I agree that most of the lesions in the prepyloric region are malignant. Again my experience coincides with Dr. Golden's, for every once in a while I come in contact with a nonmalignant lesion within the limits defined by Holmes and Hampton.

Spasm in any part of the intestinal tract is the most disturbing factor encountered and the most difficult to differentiate. Repeated examinations are often an aid, as the permanency of the defective filling is thereby directly tested. Spasm may be persistent enough to give the same deformity in filling at several examinations, and this is especially true of spasm on the gastric side of the pylorus.

In a large experience covering a long period of years I have always considered an ulcer of unusual size as "malignant until proved otherwise." Only in very exceptional cases has this rule proved wrong.

Cancer of the body of the stomach is very insidious, in many instances being inoperable when discovered.

Dr. Golden has so thoroughly covered the roentgenologic aspects of carcinoma in the lower third of the stomach that I thought it might be of interest to discuss some of the roentgen signs of cancer at the cardia. I have enumerated ten roentgen points as indicating a malignant growth in the upper end of the stomach.

It is recognized that many of these can be produced by conditions other than cancer, but if they are grouped, it is extremely probable that they will indicate whether or not the condition is malignant. The ten roentgen signs of involvement of the cardiac end of the stomach by cancer are:

1. A dilatation of the lower part of the esophagus
2. Any abnormal retention of barium in the lower part of the esophagus
3. Barium passing through the esophageal orifice in a continuous stream
4. A narrowed esophagus and an unchanging canalization through the tumor
5. A frozen mass, the infiltration preventing the normal movements of the lower part of the esophagus
6. A mass visible in a gas bubble
7. A mass visible after the first swallow of barium, with a distorted rugal pattern; a mass visible after distention of the stomach by the full meal—a contracted lumen
8. Barium forking over the mass
9. Gastric hypermotility
10. Esophageal antiperistalsis

These ten signs must be correlated with a clinical history of dysphagia, loss of weight and of appetite, regurgitation of mucus, vomiting of blood, or other positive findings.

Many patients with gastric cancer appear for diagnosis with the lesion too far advanced for surgical resection; one of the great fields for future progress in medical diagnosis is in the earlier recognition of cancer. The physician must be more ready to suspect cancer and at an earlier date. He must insist that every laboratory procedure possible be used in studying the case. To wait for the advanced typical picture of cancer is to wait too long. Before cancer of the stomach can be diagnosed early the physician must insist that roentgen examinations be made as regularly as urinalyses and repeated as often as examinations of the sputum.

The symptoms of cancer of the cardiac end of the stomach may be very indefinite even up to death. One of my patients had no complaint other than that of vomiting blood; another had only tarry stools for two weeks.

Dysphagia when present is of great value; it is found in more than half the cases. The patient may not volunteer information as to this, but answers affirmatively when directly questioned. When difficulty in swallowing or substernal pain is complained of, cancer of the cardia should be borne in mind and excluded only after thorough examination.

A cancer of the cardia involving the esophageal sphincter secondarily or by direct extension is much easier to detect roentgenographically than an involvement limited to the posterior wall. It is in this type of lesion that cancerous involvement is missed until it is so extensive that the diagnosis is obvious.

#### OPERATIVE INDICATIONS AND PROGNOSIS IN CASES OF NEOPLASM OF THE STOMACH. FORDYCE B. ST. JOHN.

I have reviewed 718 cases of carcinoma of the stomach in patients admitted to the Presbyterian Hospital, New York, in the past twenty-five years, 1908 to 1932, inclusive.

In 430 of the 718 cases an exploration was made, and in 86 per cent of these a pathologic specimen was obtained. In 212 cases, or 29.6 per cent, operation was not advised, because of advanced disease. Fifty-seven patients, or 7.9 per cent, refused operation. In 10 cases, or 1.4 per cent, the clinical diagnosis was made only at postmortem examination.

In 98 of the 430 cases explored, resection of the tumor was carried out. In other words, resection was reasonable and possible in 1 of 4.5 cases in which an

exploration was made, and in 1 of 7 cases observed in the clinic. The postoperative mortality has been steadily reduced during the twenty-five years; in the last five years it was 25 per cent.

### *Results in 98 Cases in Which Resection Was Performed*

Patients who died.....	69
Death from postoperative complications.....	33
Death from delayed postoperative complications.....	1*
Deaths at eighteen and forty-six months from pneumonia and cerebral accident, respectively, with no signs of recurrence.....	2
Deaths from recurrence.....	33
Living patients .....	26
Living (from four to twenty-one months), with lymph node involvement and showing signs of recurrence.....	4
Living (from eight to thirty-eight months) with lymph node involvement but showing no signs of recurrence.....	3
Living (from eight to forty-eight months), without lymph node involvement and showing no signs of recurrence.....	10
Living and apparently well (of 36 patients followed five years or more).....	10
Living and apparently well (of 20 patients followed ten years or more).....	7
Patients not followed.....	3

\* This patient died, two months after discharge, from peritonitis secondary to perforated jejunal ulcers. Autopsy showed no metastases or recurrence of the primary growth.

It was found from the study that the greatest factor in longevity when radical surgery was possible was the pathologic characteristic of the tumor referred to by Stout as the fungating type.

### DISCUSSION

ALLEN O. WHIPPLE: There are two points I should like to emphasize: One is the great advantage that has been found, at least in this study, in the combined efforts of the pathologist, the roentgenologist and the surgeon. I know of no more interesting subject for investigation than this group of 98 cases of stomachs resected for carcinoma. This combined pathologic, roentgenologic and clinical study indicates the type of work that is being done in clinics throughout the country.

The other point which I wish to emphasize is that the surviving patients with arrests of cancer of the stomach should be shown at least once a year in these hospital clinicopathologic conferences. One of the reasons why the impression that carcinoma of the stomach is malignant is so prevalent is that the subject is discussed at clinicopathologic conferences and very little elsewhere. The impression of the medical student, the intern and the younger clinician is largely based on cases shown at the clinicopathologic conference, and I know of nothing more healthy, from the standpoint of calling the attention of the younger clinicians to carcinoma of the stomach, than the presentation of late results, the five and ten year arrests, not only of cancer of the stomach but of other types. If that were done once a year, at least, at the clinicopathologic conference it would have a healthy and sane influence on hospital organization.

FREDERIC W. BANCROFT: I am going to confine my discussion to Dr. St. John's paper. I think it is an interesting and in many ways encouraging account of a discouraging subject. Last year, at the meeting of the American Surgical Association, Maes of New Orleans read a paper on the tragedy of gastric cancer. His report was most discouraging as a study of end-results. I think Dr. St. John has shown in this same year definite steps in progress.

I should first like to emphasize the honesty of the statistics, and particularly the statement that the deaths that have occurred while the patients were in the hospital, whether or not these deaths were due to this disease, have been reported. This has not been done in certain published compilations of statistics of operative results in the clinics of the country. I think the improvement in results which is shown by Dr. St. John to have taken place since July 1933 is astonish-

ing. It shows the value of statistics. There are many factors which may have helped to bring about this improvement. It is undoubtedly due in part to a trained group of men. It may be due in part to the use of spinal anesthesia. It may be due to the use of the Levin tube. It may be due to the efforts to prevent depletion of body fluids, and there may be other causes which have influenced the statistics. Also it occurred to me that a decade ago there was a greater number of deaths from pneumonia. I wonder whether there were deaths due to pneumonia following spinal anesthesia in the series of a decade ago as well as deaths due to inhalation anesthesia. In the statistics of the hospital with which I am associated spinal anesthetics give a higher percentage of pneumonias than any other type.

Two things are worth emphasizing in this problem of cancer of the stomach, and they also hold in cancer elsewhere. The first is preventive surgery. Surgeons have come to depend on the roentgenologist for a great deal of diagnosis and prognosis in cases involving the stomach. Perhaps they depend too much on him and do not analyze sufficiently the clinical symptoms and the general course of the disease. As long as he has them dependent on him, I think there is something else for him to do. The American Society for the Prevention of Cancer and the periodic health magazines are constantly emphasizing the necessity for periodic health examinations. A great many people come to physicians to be assured that they have not cancer. I do not think that physicians can assure those patients that they have or have not an early malignant process in the gastro-intestinal tract without some roentgenographic evidence but, at present, if a physician sends such a patient to a roentgenologist and he charges the current prices for a gastro-intestinal series, the cost of the ordinary periodic health examination becomes prohibitive. Therefore I think the roentgenologists will have to meet the physicians and say that they will give a certain type of service in periodic health examinations—for instance, a few gastric plates and one or two barium enemas—at a minimum rate. If there is a suspicion of a pathologic process there is no reason then why they should not increase the cost to the patient. I cannot see how, by doing a digital, a rectal and possibly a proctoscopic examination, with palpation of the abdomen, a physician can assure a patient that he has or has not cancer of the stomach or colon. I feel definitely sure that since physicians have adopted periodic health examinations they must carry them out and carry them out thoroughly.

The second thing which seems to me important is: What should be one's concept of a suspicious case? If the mortality following operations on gastric ulcers is considerable, and the benefit derived is the discovery of an occasional carcinoma, it seems to me one has to divide the cases into two types: (1) those of duodenal ulcer and (2) those of ulcer of the lesser curvature, which Dr. Cole and Dr. Lahey have shown will in the great majority of cases subside under rest in bed and medical therapy. Dr. Lahey has made the rule that even if there is no evidence of tumor after three weeks the patient must be sent for at the end of eight weeks and reexamined. If there is any small nicking or scar where the ulcer was the case warrants a surgical exploration. Certainly one's first concept is that rest in bed will decrease the size of the ulcer.

One then comes to the prepyloric ulcers. I am more and more convinced that if the prepyloric ulcer does not show definite roentgenologic and clinical evidence of improvement in a relatively short time the patient should be operated on. I think if one tried to follow some such concept, one might have some means of improving the five and ten year results.

LEOPOLD JACHES: I agree that early diagnosis is desirable. I must say, however, that early diagnosis is difficult if, by early carcinoma of the stomach, is meant a very small lesion which one can perhaps palpate when one has the stomach between one's fingers at the operating table and which can be diagnosed only when the pathologist has made a microscopic examination. There is perhaps one phenomenon that might help, and this Dr. Golden has referred to, namely, rigidity of the gastric wall about the area of the lesion.

When one comes to differentiate between ulcer and carcinoma, I might give another rule which is just as good as Dr. Golden's and, by the same token, does not always hold; it is that generally a carcinoma will be found in older persons and a benign lesion in younger ones. Nevertheless, Dr. Golden has shown you a film relating to a patient of over 70 who had what Dr. Golden chose to call typical evidence of carcinoma, which clinically, at least, does not seem to be that. On the other hand, once in a while one finds a very young person, even a person in the twenties, with a carcinoma of the stomach. It seems to me, therefore, that it is more important for the roentgenologist to localize the lesion than to make a positive diagnosis of a malignant or a non-malignant process. That, in the final analysis, is left to the pathologist. The size of the lesion should be an indication for the clinical procedure, whether operative or medical.

As to the disappearance of ulcers, in my experience that is a delusion and a snare. Quite a few times I have seen an ulcer demonstrated, the patient treated, and the ulcer disappear. The patient feels better. After a while he commences to have symptoms, perhaps hematemesis; he is reexamined by the same roentgenologist, and no ulcer is seen. One waits a little while. The patient does not improve and is again examined by the roentgenologist, and there is no ulcer, while clinically the case is becoming more and more desperate. The surgeon operates and finds the ulcer which was seen at the first examination, and it is practically of the same size and shape. That has happened more than once.

With regard to the differentiation of carcinoma from syphilis of the stomach, there is no typical appearance of syphilis of the stomach. The few syphilitic stomachs that I have seen have had the tumors on the greater curvature, but I doubt whether this may be considered a pathognomonic sign. The fact that a patient has had syphilis, or even the fact that he has a four plus Wassermann reaction at the time of the examination, does not mean that the defect in the gastric shadow is a gumma and not malignant.

I should like to confirm Dr. Stewart's observation that a carcinoma high up in the stomach is much more difficult to diagnose than one lower down.

In closing, I should like to take issue with Dr. Bancroft on the question of the cost or, rather, on his suggestion that the roentgenologist take just one plate "and that may be enough." Yes, that would be enough if the patient had a huge carcinoma that could be palpated through the abdomen, but if the lesion is very early, which is the one he is trying to discover by these periodic health examinations, fluoroscopy and only one plate will never help. That one plate will miss it in nine cases out of ten. I have repeatedly seen instances in which, with a series of films, the lesion was discovered on only one, and on repeated examinations the same thing happened; and although when fluoroscoping my associates and I knew what to look for, we did not find it on the fluoroscope, and the lesion which was later proved by operation to have been present showed on only one plate of a series. How is the roentgenologist or anybody else to know which one of the films is going to show the lesion?

**ROSS GOLDEN:** I did not feel that the time allotted me and the nature of the program justified a detailed discussion of roentgenologic technic. The technical points brought out by Dr. Stewart and Dr. Jaches are well taken. I may say, however, that I feel that fluoroscopy is essential, and that the patient should be examined not only upright but prone and supine and turned freely from side to side. I do not see how that can be done on films alone, and I say that without meaning to minimize the importance of films. The patient should also be examined and palpated not only when the stomach is full but when only one or two swallows of barium are trickling down. After the best positions to show the lesion have been determined fluoroscopically, the evidence can be confirmed on the films. I cannot emphasize too much that I think careful fluoroscopy is of vital importance.

I wish to thank Dr. Stewart for his ten points; many of them make the diagnosis absolutely, and others, I feel, will be present also in other conditions.

FORDYCE B. ST. JOHN: The point which Dr. Bancroft makes is important, namely, the preoperative care, including maintenance of fluid balance, rest, repeated lavage, etc. So far as postoperative pulmonary complications are concerned, I feel that no form of anesthesia can eliminate these. In my hands spinal anesthesia (*p*-n-butylaminobenzoyl dimethylamino-ethanol hydrochloride) is the most satisfactory.

## NEW YORK PATHOLOGICAL SOCIETY

PAUL KLEMPERER, *President*

*Anniversary Meeting, Jan. 25, 1934*

### CONGENITAL NEPHRITIS: SUBACUTE GLOMERULONEPHRITIS AND CARDIAC HYPER-TROPHY IN AN INFANT 4 MONTHS OLD. CHESTER R. BROWN.

On postmortem examination of a syphilitic infant 4 months old there were found contracted kidneys with finely granular surfaces and a hypertrophied heart weighing 43 Gm. Microscopy revealed subacute glomerulonephritis.

The patient was admitted to the hospital with a diagnosis of bronchopneumonia and died nine days later. The Wassermann reactions of the mother and the child were strongly positive. Neither had received antisyphilitic treatment. The delivery and pregnancy were normal.

The essential anatomic findings were as follows: The body was that of an undernourished, underweight white female child. A bloody nasal discharge was present. The heart showed marked hypertrophy, especially of the left ventricle; it weighed 43 Gm. Both kidneys were greatly enlarged, the right weighing 43 Gm. The cortex was finely granular. Punctate hemorrhages were numerous. The spleen and liver were enlarged and congested. The anatomic diagnosis was: subacute glomerulonephritis; cardiac hypertrophy; interstitial fibrosis of the lungs; chronic passive congestion of the viscera, and congenital syphilis.

*Microscopic Examination.*—All the renal glomeruli were involved but varied in size and in stage of inflammation. About 75 per cent were enlarged; the remainder were smaller and partially or completely atrophied. Bloodlessness of the loops was characteristic, although there were many old and recent capsular hemorrhages. An increase in the number of endothelial cells was present in some glomeruli; the cells of others had degenerated. Fibroblastic adhesions of the tuft and capsule, older and recent "crescents" and partial or complete hyalinization were present in various glomeruli. Some presented fibrous constricting peri-glomerulitis. Many glomeruli had disappeared. The cortical architecture was disorganized. Some tubules presented marked dilatation with epithelial fatty degeneration; many were atrophic or compressed by interstitial fibrous tissue; others showed marked regeneration of the epithelium. Numerous red cells and hyaline and granular casts filled the lumens. Small cortical scars were present, which caused focal cortical granulations. In all the vessels the media was markedly hypertrophic. In the arterioles intimal hyperplasia was marked. Necrotizing arteriolitis was present. The veins were tremendously thickened. Plasma cell infiltrations were not present, and lymphocytic foci were minimal.

In the heart, the muscle bundles were widely separated by interstitial edema, a moderately cellular connective tissue and a great number of newly formed, congested capillaries. Many of the latter had ruptured. The muscle nuclei were enlarged and stained deeply. In the larger coronary arteries and veins the media was greatly thickened. In their smaller branches intimal hyperplasia was present. All were surrounded by extensive adventitial fibrosis.

The walls of the pulmonary alveoli were moderately thickened, the capillaries were congested, and there was an increase of fibrous tissue. The lining endo-

thelial cells were increased in number, enlarged and pyknotic. Some were desquamated and contained pigment granules. The vessels were all greatly thickened with marked adventitial fibrosis. Bronchopneumonia was present.

The fibrous trabeculae of the liver and spleen were increased throughout both organs. The hepatic sinuses were congested. Erythroblastosis was absent.

In the vessels of all the viscera the media was markedly thickened.

Levaditi stains on all tissues were negative.

The microscopic diagnosis was: congenital subacute glomerulonephritis; hypertrophy of the heart; diffuse interstitial myofibrosis.

*Comment.*—The lesions in the kidney suggest that the nephritis was of longer duration than the extra-uterine life of the infant. The cardiac hypertrophy suggests that hypertension was present. Nephritis of this degree with cardiac hypertrophy has been reported in a nonsyphilitic baby 4 weeks old (Ashby). Acute diffuse glomerulonephritis has been reported (Karsner) in a full-term nonsyphilitic infant, dead fifteen minutes after birth. The fetal kidney is extremely susceptible to various toxins circulating in the maternal blood. Maternal infections, drugs and abortifacients may cause nephritis. The criteria for microscopic diagnosis of congenital syphilis vary with different authors. Lymphocytic and plasma cell infiltrations, interstitial and perivascular, are stressed by some. Others emphasize endarteritis obliterans. Only the latter was significant in this kidney. Spirochetes *per se* are probably not the direct etiologic agents in glomerulonephritis of the syphilitic new-born. The syphilitic kidney is particularly susceptible to bacterial toxins arising in the gastro-intestinal tract, which are the direct cause of the nephritis.

#### DISCUSSION

Louis Gross: When one examines the coronary arteries of an adult with hypertension one finds it very difficult to determine how much of the intimal lesion is due to the hypertension and how much to the changes belonging to the age. After the first decade, or at times even before the end of the first decade, some coronary vessels may show changes which resemble in their extent those found in vessels of much older persons. Dr. Brown's case is a very fortunate one from this point of view; here is an infant 4 months old in whom the intima was undoubtedly a simple structure. Whatever changes may have taken place, one may reasonably assume that they were due to the hypertension that must have been present, as evidenced by the thickness of the myocardium. Just what did the intima of the coronary arteries look like?

Paul Klemperer: It is not safe to determine the duration of nephritis from the histologic appearance. Sometimes nephritis may proceed very rapidly, and a picture which it is customary to correlate with a few months may obtain within a shorter period. I wonder, therefore, whether one can definitely say that this is a case of congenital nephritis. Four months is, after all, a period the changes of which in an adult correspond with the picture that is seen here. The changes might proceed much more rapidly in the infant, and the same picture might be produced in a shorter time.

Chester R. Brown: In the smaller coronary vessels and in some of the larger ones the intima was hyperplastic.

It is true that this advanced nephritis might be produced by a glomerular inflammatory process originating after the birth of the infant.

AN ATYPICAL CASE OF MULTIPLE MYELOMA. SHELDON A. JACOBSON and MARTIN G. VORHAUS (by invitation).

The case presented seems to be of interest from both a theoretical and a practical point of view. A. R., 42 years old, was first admitted to the hospital on June 20, 1932. She had been under intermittent treatment in the clinic of the hospital for four years with symptoms involving almost every system of her body. For some time during this and subsequent stays in the hospital the same confusing

multiplicity of symptoms continued. With time, however, the following signs and symptoms assumed particular prominence both to the clinician observing her and in view of subsequent observations at autopsy.

There was marked, general, progressive muscular weakness. General pain was complained of (the lower part of the back being most troublesome), and gastrointestinal distress made its appearance. The musculature and subcutaneous tissues were of a putty-like consistency. Translucent, waxy papillomas appeared on the eyelids, lips, buccal mucosae and anus. The vertebrae and the sternum were sensitive to palpation. Between 6 and 8 Gm. of Bence-Jones protein was eliminated daily. There was moderate anemia. The white blood count varied between 11,500 and 20,600; the differential count was normal. (A few days before death the white blood count reached 27,900). Repeated careful roentgen examinations of the skeleton revealed no abdominal conditions.

The diagnosis was held to lie between multiple myeloma and idiopathic amyloidosis.

At autopsy removal and sagittal section of the vertebral column and careful examination revealed nothing noteworthy in the gross. The marrow of the ribs, on the other hand, was suspiciously pale and gelatinous. The musculature of the back had completely lost its normal color, being deep yellow. The tongue was enlarged. Microscopic section showed myeloma cells infiltrating all parts of the spongiosa of all vertebrae and ribs examined. An amyloid-like substance was present in the lungs, liver, spleen, stomach, intestines, tongue, bladder, uterus, fallopian tubes, lymph nodes and skin. Chemical examination of the muscles failed to reveal appreciable quantities of amyloid.

The chemical derangements which characterized the local and general metabolic processes of this patient would no doubt shed much light on the nature of myeloma, could they be properly interpreted. That is beyond our power at present. Of immediate practical importance is this new demonstration of the insufficiently appreciated fact that a pronounced myelomatous involvement may fail to give roentgenologic evidence of its existence. Even a careful gross examination of affected bone may mislead a pathologist into excluding a myeloma which is actually present. Doubt is thrown, therefore, on the existence of the so-called one-bone myeloma and on the existence of essential amyloidosis with Bence-Jones proteinuria.

#### DISCUSSION

JACOB FURTH: Since tumors were absent, and the disease process was diffuse and not "multiple," would you not prefer designating the condition as myelomatosis? What are, in your opinion, the cells that infiltrated the bone marrow?

MENDEL JACOBI: I should like to ask Dr. Jacobson whether or not there was any evidence of cellular necrosis in the many slides which he unquestionably studied, and then I should like to say a word about giant cells in amyloidosis, a subject which Dr. Jacobson brought up. Experimentally my associates and I have found (and we have been working experimentally with amyloidosis for a considerable period of time) that in the very earlier stages of amyloidosis, when it is intracellular rather than extracellular, there is often a giant cell reaction. As the extracellular amyloid becomes manifest and increases in amount there occur simultaneously necrobiotic changes in the giant cells, so that in places we can no longer distinguish the cell outlines; the further the process progresses, the fewer are the intact giant cells and the more numerous are the fragmented giant cells and giant cell nuclei present in the amyloid masses. Finally, as the amyloid replaces large areas of parenchyma, all evidences of cellular reactivity vanish. These changes my collaborators and I have described in detail in a recent number of the ARCHIVES (17:50, 1934).

I should like to ask whether there was any amyloid in the kidney. I presume there was not. It is interesting that in the larger number of patients with Bence-Jones albuminuria amyloid is not found in the kidney. The Bence-Jones protein

may be the matrix of the amyloid, and it is likely that its secretion and excretion through the kidneys do not permit a deposition of amyloid in this organ.

PAUL KLEMPERER: I am not quite clear on the diagnosis of myeloma in this case. I feel rather disinclined to believe in the so-called diffuse myeloma. But, granted that such an entity exists, there is one question I should like to ask: What are these cells which Dr. Jacobson calls myeloma cells? Rustitzky, in his definition, maintained that the hyperplastic nodules in the bone marrow consist of nothing else than what is found in the bone marrow. If these cells are not bone marrow cells, what are they? Dr. Jacobson mentioned extramedullary myeloma without involvement of bone. I do not understand what this is. I think one might not be too strict and too orthodox if one believes that a myeloma must be absolutely limited to the bone. A myeloma without skeletal involvement I cannot understand. I might have misunderstood Dr. Jacobson, but I think he mentioned cases in which "myeloma cells" were found without skeletal involvement.

In regard to the question of the lymphocytic origin, I might also have misunderstood Dr. Jacobson when he said that his concept contradicts that of the lymphogenic nature of the myeloma cells. I think there are cases of myeloma on record in which myelocytes were found. The myeloma cell, or the cell which composes the nodules in the bone, need not be of lymphocytic origin. I do believe the cell in the so-called plasma cell myeloma is not a true plasma cell, but an atypical lymphoblast or, better, a hemocytoblast. While it is probable that the greater number of myelomas do not consist of mature and typical bone marrow cells, there are certainly some which do consist of them. It is true that one cannot always identify the cells of a myeloma with a definite, mature bone marrow cell, but one can compare them with the ancestral cell of the bone marrow cell. I should like to know what Dr. Jacobson calls myeloma cells, and what is the difference between his concept and the classic concept of myeloma.

SHELDON A. JACOBSON: The question of the origin of myeloma cells is a pitfall for the feet of the unwary, and I have great hesitation in speaking of that and in defining a myeloma. Since, however, I am forced to answer that question, I should say that my diagnosis of a myeloma, at least of an intraskeletal myeloma, for the moment, is based on the presence of a tumor mass within the bone or soft tissue, composed preferably of cells of the type that are called plasma cells—like marrow cells, large cells with abundant cytoplasm and eccentric nuclei, containing chromatin divided up into many little points. I have made the diagnosis in this case on that basis. That was the preponderant cell type in the smear which I made of this marrow; it was a cell not native to the bone marrow. It did not belong to the red cell series or to the myelocytic series. It was not a reticulum cell. It did not belong to the lymphocyte cells. It had the closest identity with the sort of cell that is found in some atypical cases of myeloma in which one finds nodules of soft tissue in great numbers growing in and destroying the bone marrow. Such tumors may be accompanied by Bence-Jones proteinuria. That is the best definition I can give.

The origin of the cells is not yet settled, and I hardly care to make a statement. I think that the best discussion is that of Wallgren, who relates the primitive cells in some way with the myeloid series. I think they are stem marrow cells which have not yet differentiated.

Cases have been reported in which the tumors did not involve the skeleton at any point. There was a case of a large tumor of the nasopharynx which was made up of just the cells which I have described, such a growth as one is accustomed to call plasma cell myeloma. It was associated with amyloidosis and Bence-Jones proteinuria. If these tumors can grow outside the skeleton and arise in lymphoid tissue, as this nasopharyngeal one is reported to have done, the myeloma cell must be a cell that is sometimes, at least, derived from lymphoblastic tissue. Dr. Klemperer throws doubt on the freedom of the skeleton from myeloma in such cases. In agreement with him, I point out that mine is a case which on microscopic examination showed a predominant cell type which one calls the myeloma

cell, but which had, nevertheless, nothing to be seen on macroscopic examination. How much more difficult it is, therefore, to rule out the presence of a skeletal myeloma when one has a more limited autopsy, and sometimes not an autopsy, but just a roentgen study! I think I have answered the first speaker as well as I can.

In reply to Dr. Jacobi, I wish to say that I did not observe any necrosis in this tumor. The predominant feature was degeneration. Giant cells were rare, though well defined. I saw no signs of giant cells fading out or merging into the amyloid which surrounded them. In all of them the nuclei were perfectly dark. It is my impression that most of the cells had nuclei which were likewise dark and well defined.

#### STRUCTURE OF THE NORMAL CALVARIUM IN DIFFERENT AGE GROUPS. SIDNEY A. BERNSTEIN (by invitation).

A detailed microscopic examination was made of calvaria ranging from that of a 7 months premature infant to that of a man aged 82 years. These were arbitrarily divided into four groups:

Group 1, those up to 1 year of age, representing the infantile calvarium.

Group 2, those from 1 to 20 years of age, representing the period of greatest growth.

Group 3, those from 20 to 50 years of age, in which the greatest change in the internal architecture was taking place.

Group 4, those from 50 to 80 years of age, presenting senile changes.

The infantile calvarium (thickness of 0.8 mm.) presents no division into two compact tables with a spongiosa between. Instead the bone is arranged in overlapping sheets, like shingles, composed of (1) immature intramembranous bone, which is formed at the suture, and (2) a somewhat riper bone, the result of pericranial apposition. The former type diminishes on receding from the suture. The preformed connective tissue becomes incorporated in the premature bone as long Sharpey fibers. Growth takes place at the suture in a manner analogous to that at the epiphysis of a long bone; that is, the connective tissue proliferates at the center, while becoming ossified at the margins of the suture. Four important growth changes are taking place: (1) The skull is enlarging in its transverse plane; (2) it is enlarging in its vertical plane; (3) it is getting thicker; (4) not only the skull as a whole, but each element of it, is moving in a vertical direction.

The skulls in group 2 have a thickness of from 1 to 6.5 mm. A differentiation has taken place into tabula externa, diploe and tabula interna. The compact tables consist of bone laid down by the dura and pericranium, which is lamellated horizontally. Early this apposition takes place without interruption; therefore no cement lines are present, but after from five to seven years this process slows down and cement lines appear. At this time, also, the older and more centrally placed bone begins to undergo creeping resorption, and haversian systems make their appearance. The tabula externa and the tabula interna are similar in structure, except that the former is thicker and the change is more advanced. The cellular lymphoid marrow of the diploic spaces begins to show islands of fatty marrow.

Group 3 have a thickness of from 5 to 8 mm. Haversian systems are present in all layers of the compacta. This remodeling may be so far advanced, especially in the tabula externa, as to wipe out even the last vestiges of the original horizontal lamellated bone. Owing to resorption on the pericranial surface of the tabula externa and apposition of bone on its endosteal surface, this table may move centripetally or, more rarely, the changes may be reversed and the table move centrifugally.

Group 4 have a thickness approximately the same as in the foregoing period. The diploe encroaches on both compact tables as the result of the resorption of bone on their endosteal surfaces. These tables are now thin and porosized. The diploic spaces are large and the trabeculae thin with little knobs of localized

apposition of bone here and there. Rests of horizontally lamellated bone are found deep in the diploic trabeculae as proof that these occupy a niveau which was formerly part of the compact table. The bone cells of the older bones die off and show absence of nuclear staining.

#### DISCUSSION

ALFRED PLAUT: Up to the age of about 8 years the dura mater at the convexity is firmly adherent, as it is at the base throughout life. After that age the dura mater is separated from the bone at the convexity. What is the histologic basis of this difference?

DAVID P. SEECOF: May I add that in senility the dura again becomes adherent to the under surface of the bone. Why?

I think this is a remarkable paper. There is need of more studies of this kind. I should like to ask how many cases there were in each group, in order that one may have an idea of how far the normal variations have been covered.

I should like to ask whether concomitant with the changes in the bone there were changes in the blood supply. That is important in relation to the changes that occur in bones in diseased states, such as osteomyelitis. I think from what I can gather that there must be changes in the blood supply, especially in the venous supply to the diploe.

H. L. JAFFE: Dr. Bernstein carried out these interesting studies in Vienna, in Erdheim's laboratory, when he was a fellow from the Hospital for Joint Diseases.

One is prone to consider the calvarium as something that has to be got out of the way in approaching the cranial cavity. After it is removed it is usually casually examined, then put down and forgotten. The calvarium, if it is studied, presents tremendous normal variation. I had occasion to find out something about the normal calvarium. I wanted to make a comparative study of the normal calvarium in connection with some calvaria which I was studying. When I looked into what was known about the normal and the pathologic anatomy of the calvarium I found a large hiatus in the literature. There was considerable information on the embryologic development of the skull, especially the calvarium. There was a good deal of information on suture closure and suture function. The anthropologic aspects of the skull and the comparative anatomic details were well known. But when I came to getting practical information on how thick the calvarium is in a man 60 years of age I found that search of the literature was of little avail. Furthermore, it is difficult to learn exactly the measurements of the outer and inner tables and of the diploic zone. Dr. Bernstein has done much to fill in the gap in the information. He cautiously states that all the skulls that he studied were so-called normal skulls, and he said that they were so-called normal because there was no cranial disease. Not infrequently one comes across a heavy, sclerotic skull in a very old person. There is really little information as to why skulls sclerose with advancing age. In connection with senility, Dr. Bernstein states that eccentric atrophy may come with advancing age. This, the symmetrical osteoporosis of old age, leads to erosion of the outer table in the parietal region. With more knowledge of the normal, it would be simpler to understand the basis for the thickening of the skull in Paget's disease and the details of the changes in other fibrotic diseases.

SIDNEY A. BERNSTEIN: In answer to Dr. Plaut's question regarding the separation of the dura, I think the matter depends on whether bone is being apposed or resorbed on the surface. When it is being apposed, a thick cambium layer is present and adherent. The fibers of the periosteum are being incorporated in the newly formed bone as Sharpey's fibers. As a result, the dura is adherent to the bone and is separated with difficulty. But when resorption of bone is taking place, the Sharpey fibers together with the surrounding bone disappear, and thus the connection between the periosteum and the bone is loosened.

The question was raised, Why should the dura be adherent in advanced age? In many of my calvaria of about the age of 40 to 50, especially toward the

parietal region, I found a different type of bone, which I did not mention for lack of time. This bone was laid down on the outer surface of the tabula externa in a very thick layer, was very primitive, taking on an extremely dark blue staining reaction, and consisted of confluent calcareous granules. Over this bone was a very thick layer of osteoid tissue, and running through both these layers from the periosteum were many Sharpey's fibers. This may explain the renewed adherence of the periosteum to the bone at a time when the rule otherwise is bone resorption on the surface.

As to Dr. Seecof's question concerning the number of cases in each group, I made it a point to take at least three cases from each decade, so when the group, e. g., group 3, encompasses three decades, nine calvaria were studied. From each skull cap three sections were taken from a wedge beginning at one side of the sagittal suture and extending across the midline to the opposite temporal region, including the suture between the temporal and parietal bones.

As regards changes in the blood supply, both in the diploic spaces and in the compact tables, I found much more vascularity in the younger age groups. In the very young the compacta contained many Volkmann's canals, especially the tabula interna. As mentioned, the contents of the diploic spaces changed with oncoming age from a vascular marrow to a cellular marrow and finally to a fatty marrow. It may be that this decrease in blood supply is the cause of the frequent occurrence of dead cells in aged skulls. Bone cells were found in which the nuclei took no stain, and this not only in the horizontal lamellated bone, but also in the older haversian systems. I say "older" because one can fairly accurately judge how old they are by the difference in the staining reaction; the bone in the older haversian systems takes a more basophilic stain than does that in the younger ones.

#### PERIARTERITIS NODOSA (NECROTIZING ARTERITIS), WITH A NOTE ON ABDOMINAL RHEUMATISM. CHARLES K. FRIEBERG (by invitation) and LOUIS GROSS.

In the last two years at Mount Sinai Hospital there have been eight patients with necrotizing inflammatory arterial lesions (periarteritis nodosa) whose condition was found at autopsy. Of these, four had unquestionable rheumatic heart disease, including Aschoff bodies in the myocardium. Strict criteria were employed for both the diagnosis of rheumatic heart disease and that of periarteritis nodosa. The clinical basis for the diagnosis of rheumatic heart disease consisted of acute polyarthritis with fever, pericarditis and signs of valvular disease. Pathologically there were characteristic deformities of the valves, auricular lesions, pericarditis and Aschoff bodies in the myocardium in each case. The diagnosis of periarteritis nodosa was not based merely on isolated instances of necrotizing arteritis in single organs. The lesions were very extensive, involving at least the heart and the kidney in each case and generally numerous other organs as well. The lesions were grossly visible either as markedly thickened vessels or as characteristic nodules along the course of the blood vessels. Thromboses, infarctions and aneurysms were present. Microscopically there were panarteritis with necrosis of the arterial wall, periarterial inflammation, thickening of the intima, narrowing of the lumen and occasionally evidences of healing.

Furthermore, the lesions were severe enough to give rise to clinical symptoms. Orchitis was present in both the males. Abdominal pain was present in all four patients, and in two was sufficiently severe to lead to operative intervention. Symptoms of renal involvement were present in all. Had we been less strict in our criteria of heart disease or of periarteritis nodosa, we should have had a considerable number of additional cases available besides the four of combined rheumatic heart disease and periarteritis nodosa which we present. (The case reports with slides were presented.)

A relationship between periarteritis nodosa and rheumatic fever has been repeatedly conjectured. We do not believe that the association has ever been conclusively demonstrated. The relationship has been suggested by certain similarities

in clinical features, by the suggested allergic nature of the diseases and by occasional instances of arthritis in patients with periarteritis nodosa. However, in the latter instances only rarely has mention been made of the finding of endocarditis at autopsy and never have Aschoff bodies been described in these cases.

Further evidence for the relationship under discussion has been found in the vascular lesions described in rheumatic fever. Aschoff, Geipel and Coombs described necrotizing inflammatory lesions in the finer branches of the coronary arteries in rheumatic heart disease. Klotz described vascular lesions in rheumatism which were examples rather of productive than of destructive inflammation. Watjen described a panarteritis with destructive changes involving the smaller coronary branches. In all of these cases, however, the pathologic alterations were present only in the heart or in the heart and aorta. More significant is the report of Von Glahn and Pappenheimer who found vascular lesions with necrosis and inflammation involving many organs in ten of forty-seven cases of rheumatic heart disease. However, these lesions were present in only one or two organs in any given case. They were found only on microscopic examination. They apparently did not give rise to clinical symptoms. Thromboses, aneurysms and infarctions were not present. The authors themselves believed that there were clearcut differences between these lesions and those of periarteritis nodosa.

In the cases which we have described not only was there an undoubted association of rheumatic heart disease and periarteritis nodosa at postmortem examination, but also in the clinical course of the disease in three of the four cases the symptoms generally ascribed to rheumatic fever were present simultaneously with those generally ascribed to periarteritis nodosa. We therefore suggest that the vascular lesions termed "periarteritis nodosa" may be caused, among other conditions, by rheumatic fever. Furthermore, it is suggested that in cases of acute rheumatic fever with severe abdominal symptoms sometimes leading to exploratory operations, an organic basis for the symptoms may be found in the vascular lesions of periarteritis nodosa.

#### DISCUSSION

C. J. Sutro: As subcutaneous nodules are present both in rheumatic fever and in periarteritis nodosa, the presence of both types in one case would be most unusual. Were there any subcutaneous nodules in these four cases? If there were, were they removed, and if so, what did they show?

IRVING GRAEF: Two cases bearing on this subject have been studied recently at Bellevue Hospital. They raised the question, certainly in my own mind, as to the possible identity of the lesions in the blood vessels with those of the rheumatic pattern. One was that of a man, 65 years of age, who had inactive rheumatic heart disease and who died in heart failure with severe mitral stenosis and auricular thrombosis. Clinically several lesions were noted beneath the skin on his back and once on his left arm; they had a dusky blue tinge and disappeared. They were thought to be subcutaneous nodules, but on removing one of them a lesion characteristic of periarteritis nodosa was found. At necropsy four more lesions were found, one along the aorta, one in the bladder and one in each kidney. As I said before, the heart was entirely negative for *active* rheumatic carditis. The man died of heart failure, and whatever factor produces periarteritis nodosa produced these lesions.

The second case was that of a 15 year old boy who had an acute respiratory infection with an exanthem, and died in uremic coma with convulsions three weeks later. He was in the hospital only twenty-four hours, so the clinical data are inadequate. At autopsy he had widespread necrotizing arteritis with almost universal involvement of the renal arterioles—arteriolitis. There was an acute necrotizing nonbacterial valvulitis (with no gross lesions on the valves) and no Aschoff bodies in the myocardium. This case I think belongs to the same group as Lamb's case described in 1912-1913.

I believe the lesions in periarteritis-nodosa are certainly distinct, and in many cases can be sharply separated histologically from the lesions of the blood vessels

in rheumatic carditis, as one sees them in the common run of cases, but I am not sure that even an identical appearance points to an identical etiology, even when rheumatic carditis is present.

The wide base from which necrotizing arterial lesions can spring seems to be widening every year. I need only recall the case reported by Dr. Helpern and Dr. Trubek of a person with gonococcal endocarditis in the right side of the heart, sepsis, and necrosing arteritis systemically distributed. Similar lesions have been reported in other acute infections (I have seen them associated with pneumococcic infections), tropical diseases like yellow fever, typhoid and so on.

CHARLES K. FRIEDBERG: Cases in which subcutaneous nodules are found are always very helpful when they appear clinically. In none of these cases were such nodules discovered. Of course, as I have pointed out, the diagnosis of periarteritis nodosa has sometimes been made when they were not present.

In regard to Dr. Graef's interesting cases, I think that the evidence for the etiologic relationship between the vascular lesions which we observed and the rheumatic fever is not based only on the high incidence of rheumatic fever in our series of cases of periarteritis nodosa. In certain of the cases the period of the symptoms was very brief, and some of the symptoms were definitely those of rheumatic fever, and others, such as orchitis, were definitely those associated with periarteritis nodosa. Furthermore, there were evidences of nephritis and uremia, which are so uncommon in uncomplicated rheumatic fever (Baehr and Schiffrin) that we can hardly believe them to have been a part of the rheumatic fever except so far as rheumatic fever and periarteritis nodosa are part of one and the same complex. Abdominal pain is a more frequent, though not common, symptom of rheumatic fever, but then again abdominal pain is one of the triad of symptoms which appear so commonly in periarteritis nodosa that I feel one is not going very far afield in concluding that in these cases rheumatic fever was a definite etiologic factor in the arterial lesions. I should like to add that we have had other patients with histories of rheumatic fever, evidences of valvular lesions and thrombo-endocarditis of the valves without Aschoff bodies whom we have not included in the series but whose condition may very well be part of a general rheumatic infection.

LOUIS GROSS: I should like to say a few words in regard to Dr. Graef's remarks. I should like to turn the tables around and say that I am not convinced that these two cases are not cases of rheumatic fever, but it was just because we had this doubt in our mind that we leaned over backward and included those cases which gave us only convincing evidence, namely, the Aschoff bodies. We do not want to leave the impression in the mind of any one that we consider periarteritis nodosa as perhaps largely occurring in rheumatic fever. Undoubtedly there are a great many cases of periarteritis nodosa in which rheumatic fever is not associated, and there are many diseases causing cardiac lesions, but we want to point out that among the diseases in which both conditions occur, rheumatic fever is one.

## Book Reviews

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**The Lyophilic Colloids (Their Theory and Practice).** By Martin H. Fischer, Professor of Physiology, the University of Cincinnati, and Marian O. Hooker, Research Associate in Physiology, the University of Cincinnati. Price, \$4.50. Pp. 250, with 84 illustrations. Springfield, Ill.: Charles C. Thomas, Publisher, 1934.

This book presents a summary of the work of Fischer's laboratory during the past fifteen years. It is divided into three parts. The first part records in detail certain experiments which illustrate the general nature of the lyophilic (hydrophilic) colloids. The second part deals with chemical applications, and the third with biologic applications. The physician and physiologist will be interested primarily in the third part.

The authors point out that when certain colloid systems are cooled, for example, phenol/water, quinoline/water, soap/water, gelatin/water and casein/water, the nature of the solution gradually changes from one of X-in-solvent to one of solvent-in-X, with many intermediate types of solutions, all of which have different chemical and physical properties. Experiments are recorded in detail which show how these properties are affected by the addition of various substances. Particular attention is paid to the effect of acids and alkalis on the hydration capacity of the solid phases of such systems. The theme is essentially that the properties of protoplasm resemble more closely those of that phase of a colloid system in which water is dissolved in X than those of the phase in which X is dissolved in water. The laws of the dilute solution apply to the second type of solution, but not to the first. The authors believe that many erroneous deductions have been made as a result of trying to apply these laws to protoplasm. According to them, this concept explains why "no physical chemist has ever found any direct or simple relation between any one of the properties of the dilute solution phase of a lyophilic colloid system (its  $p_H$ , its osmotic concentration or its electrical properties) and the 'behavior' of the total system." Their disagreement is chiefly with the physical chemist. In their opinion, he degrades the cell to a drop of water in which various salts and nonelectrolytes are dissolved. He relegates the proteins to a third place and mentions the fats and carbohydrates as an afterthought. "To get a 'behavior' out of" the mixture "at all comparable with that of a living cell" he surrounds it with a "membrane," the properties of which are said to vary in different types of cells. "The colloid chemist" [Fischer] "thinks most of these things wrong. . . . He begins by wiping out the cell 'membrane' of the physical chemist because entirely hypothetical. With the pure chemists, he places the proteins of the living mass first. He mentions next the salts, but not as things which are merely mixed into the protein but as materials which, as acids or bases, were originally united with the protein. Third, he puts the water, but not as a solvent for the protein-salt complex but as a material dissolved in the latter. This membraneless hydrated protein-salt compound is the unit of his living mass. Into it (for the present) he merely mixes (emulsifies) the fats and the higher carbohydrates which are found in cells." The authors state that there are difficulties in the osmotic concept of the cell. On the biologic side, they claim that "an osmotically constructed living cell is an impossibility." On the physicochemical side, they state that the so-called "quantitative" experiments failed. They claim that the exchange of water between cells and their environment depends on the hydration capacity of colloids, which in turn is affected by various substances, notably acids and alkalis. The pressure theory of edema "breaks of its own weight," and edema becomes a "problem in colloid chemistry. . . . We have to look to changes in the tissues and cells themselves for the first 'causes' of an edema."

The book contains much repetition, but may be read with profit by all those interested in biology and physiology, particularly those who are interested in the exchange of water between cells and their environment. The experiments on various colloid systems are of interest, but the interpretation of various biologic phenomena by analogy from them has been carried too far. While the hydration capacity of colloids may play a rôle in the exchange which occurs between cells and their environment, the authors' attempt to establish it as the only factor will meet with opposition. Physical chemists will find it difficult to believe that they have satisfactorily excluded osmosis and that pressure has been adequately disposed of as a factor in cardiac edema. Many chemists will question the authors' theme, namely, that protoplasm is a solution of water-in-X. Nevertheless, the book stimulates thought about fundamental biologic problems and, is therefore, a valuable contribution to physiology.

## Books Received

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LA SYPHILIS EXPÉRIMENTALE. ÉTUDE CRITIQUE ET NOUVELLES RECHERCHES. P. Gastinel, Professor agrégé, and R. Pulvénis, Chef de Laboratoire, à la Faculté de Médecine de Paris. Price, 45 francs. Pp. 244, with 19 figures and 4 plates. Paris: Masson et Cie, 1934.

This is a critical review of the work on experimental syphilis with results of new investigations. The first chapter traces the early stages in the study of experimental syphilis in man and animals. Then come chapters on: experimental syphilis in apes, monkeys, the rabbit, and other animals; on the routes of generalization, the infectiousness of the lesions, and of the different organs; on experimental syphilis in general and the factors that may modify its evolution; on unapparent experimental syphilis; on immunity in experimental syphilis, and on the reaction of Meinecke in syphilitic rabbits. The monograph will be of much interest and value to all who are concerned in the study of experimental syphilis.

JOURNAL OF TECHNICAL METHODS AND BULLETIN OF THE INTERNATIONAL ASSOCIATION OF MEDICAL MUSEUMS No. XIII. Maude E. Abbott, M.D., editor. Price, \$2.00. Pp. 204. Montreal (3640 University Street): International Association of Medical Museums, 1934.

This number contains: a sketch of D. S. Lamb (1843-1929) of the Army Medical Museum in Washington; editorials on various topics; articles on museum administration, museum and autopsy technic, photographic methods, microscopic technic, teratology, cardiovascular anomalies and endometrial hyperplasia; book reviews; obituaries of international members; proceedings of the association, the constitution, and a list of the members.

BRUCELLA INFECTIONS IN ANIMALS AND MAN. METHODS OF LABORATORY DIAGNOSIS. I. Forest Huddleson, Department of Bacteriology and Hygiene, Michigan State College. Price, \$2.25. Pp. 125, illustrated. New York: Commonwealth Fund, 1934.

DIE PATHOLOGISCHE-ANATOMISCHEN GRUNDLAGEN DER CHIRURGIE DES REKTUMKARZINOMS. Von Priv.-Doz. Dr. Heinrich Westhues, Erster Oberarzt der chirurgischen Universitäts-Klinik Erlangen. Mit einem Geleitwort von Prof. Dr. Schmieden, Frankfurt a.M. Price, 29.50 marks. Pp. 113, mit 107 zum teil farbigen abbildungen. Leipzig: Georg Thieme, 1934.

# ARCHIVES OF PATHOLOGY

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## THE RENAL GLOMERULUS IN VARIOUS FORMS OF NEPHROSIS

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The desire to correlate anatomic with physiologic changes has been one of the driving forces in the advancement of knowledge. It has gradually led to the study of minute phenomena either through closer and more prolonged observation or through the introduction of newer methods allowing the discovery of new facts. These comments apply in particular to recent efforts which have been extended in regard to histologic changes occurring in the kidney and especially in the glomerulus in various types of renal disease. The application to the kidney of the stain combining Mallory's aniline blue and Heidenhain's azan carmine as described by McGregor<sup>1</sup> has opened an entirely new approach to study of the structure of the normal glomerulus, and it has brought out some important considerations, not only of glomerular structure, but also of the pathogenesis of glomerular disease. Studies have recently been published<sup>2</sup> regarding the glomerulus in the normal kidney of man in lipoid nephrosis, clinical glomerular nephritis, hypertension, eclampsia and subacute bacterial endocarditis. The present article will deal with my observations of the glomerulus in cases of simple nephrosis.

Nephrosis is generally considered to be a condition in which there are degenerative renal changes in contradistinction to inflammatory or nephritic and vascular or nephrosclerotic changes. Some use the term "tubular nephritis" or "parenchymatous nephritis" to designate the condition which others call "nephrosis." Cases of nephrosis are essentially in two main groups: (1) that of the so-called lipoid or chronic nephrosis and (2) that of simple nephrosis. Lipoid or chronic nephrosis is probably a distinct renal disease, often associated with chronic glomerular nephritis, and bears no evident relationship to the various types of simple nephrosis. The latter is the result of the action on the kidney of some abnormal substance producing degenerative changes of varying degree, and the name assumed for the individual type is frequently that

Work done in the Section on Pathologic Anatomy, the Mayo Clinic.

1. McGregor, Leone: Am. J. Path. 5:545, 1929.

2. Wilbur, D. L.: Arch. Path. 12:413, 1931.

of the supposed active agent; that is, bile nephrosis, chemical or corrosive mercuric chloride nephrosis, etc. In view of the paucity of knowledge, classifications of these conditions, whether based on etiologic, clinical or pathologic grounds, are highly controversial, probably inaccurate and subject to revision. However, for purposes of study it is helpful temporarily to classify these cases according to types. In the present study, the following types have been recognized: (1) acute simple or toxic nephrosis; (2) bile nephrosis; (3) chemical nephrosis; (4) renal changes of pregnancy and eclampsia.

Studies of the glomerulus in cases of lipoid nephrosis will be considered separately.

Before the observations are considered in detail, attention should be directed to several facts. In the first place, there may be considerable debate as to the primary or secondary nature of the renal lesion in cases of simple nephrosis. This question is of great importance. In the second place, it should be recalled that the glomerulus is but one part of the anatomic and functional unit of the kidney. Although for purposes of anatomic and physiologic study it is profitable to consider the glomerulus separately because of its different structure and function, care must be exercised in drawing too many sweeping conclusions from the study of a part of the unit only. Last, it should be pointed out that visible anatomic and physiologic changes may not always coincide and that therefore an organ which appears normal anatomically may have been functioning abnormally, and vice versa.

#### MATERIAL FOR STUDY

The selection of the cases for this study of glomerular characteristics in nephrosis requires some explanation. It was possible to obtain cases by two methods. First, the selection could be made on the basis of clinical data and diagnosis only; second, it could be made on the basis of the pathologic diagnosis. The latter method of choosing cases was followed because it seemed more likely to include the type of cases wanted, particularly since the pathologic diagnoses were made with knowledge of the clinical findings. Consequently the records of the Section on Pathologic Anatomy of the Mayo Clinic from 1923 to 1931 inclusive were searched, and all cases in which a diagnosis of nephrosis or its approximate equivalent had been made were included in the study. The majority of the cases may be considered as examples of acute simple or toxic nephrosis or of terminal nephrosis. Many of them occurred in patients who had been operated on and in whom, postoperatively, such complications as infections and gastro-intestinal obstruction or retention developed.

The following modified method<sup>3</sup> of using the Mallory-Heidenhain stain proved very satisfactory for routine work.

Take tissues fixed in formaldehyde solution (if the tissue is old, use the ammonia bath for one hour). Make paraffin sections 4 or 5 microns thick. Dry the sections in the oven at 37 C. over night. Remove the paraffin from the sections and wash them in water; then put them in Zenker's solution for one hour. Wash them in tap water for ten minutes, remove the crystals with compound solution of iodine and clear the sections in 2 per cent sodium hyposulphite and water.

1. Place the sections for four hours in a first Weigert mordant made as follows: potassium bichromate, 5 Gm.; fluorochrome, 2 Gm., and distilled water, 100 cc., boiled together.

2. Wash the sections in tap water and place for two hours in a second Weigert mordant made as follows: acetate of copper, 5 Gm.; fluorochrome, 2 Gm., and distilled water, 100 cc., boiled together.

3. When the mixture is cold, acetic acid (36 per cent), 5 cc., and diluted formaldehyde solution, 10 cc., are added.

If the tissue is old, replace the sections in the ammonia bath for two minutes. Wash for one hour in running tap water.

4. Place for forty minutes in azan carmine, prepared as follows: Put 1 Gm. of azan carmine in 100 cc. of distilled water, heat, cool, filter at room temperature and add 1 cc. of glacial acetic acid.

5. Differentiate in aniline alcohol (watch under the microscope). For aniline alcohol add 1 cc. of aniline oil to 100 cc. of 95 per cent alcohol.

6. Remove aniline with acid alcohol and wash in water quickly.

7. Place for two hours in 5 per cent phosphotungstic acid. Wash quickly in water.

8. Place for from ten to twenty minutes in Mallory's aniline blue and orange G without fuchsin. This is made by mixing together aniline blue, 0.5 Gm.; orange G, 2 Gm., and distilled water, 100 cc., and then adding acetic acid.

9. Wash quickly in water and differentiate in absolute alcohol (watch under the microscope).

10. Treat with xylene and balsam and mount.

Studies of the normal renal glomerulus by this method have already been published and serve as a basis for the present observations.

#### I. ACUTE SIMPLE NEPHROSIS

Acute simple nephrosis, a type of that group 2, previously defined, which is designated by the term simple nephrosis, comprises the majority of cases of the so-called nephroses. Acute simple nephrosis is known also as toxic nephrosis, terminal nephrosis, cloudy swelling, acute tubular nephritis, acute parenchymatous nephritis and so forth. A strict limitation of this type is naturally impossible, for it depends on whether one is speaking in terms of pathologic changes or in terms of clinical obser-

3. de Galantha, Elena: Personal communication to the author.

vations. One can define it perhaps as a disturbance, usually of short duration, with primarily degenerative changes in the renal tissue, principally the tubules, manifested clinically by albuminuria and occasionally by cylindruria, the result of a state of fever or so-called toxemia. It should be emphasized that the renal disease is generally but one expression of a widespread so-called toxic effect, and that other organs may be and usually are likewise affected. Consequently, the renal changes are secondary, and the disease in the kidney is not primarily a renal disease, such as is suggested in a case of chronic glomerular nephritis. Another point to be stressed is that so far as the lesion in the kidney is concerned it is not necessarily irreversible and consequently it is one from which clinical recovery can occur. This fact in itself distinguishes acute simple nephrosis from acute nephritis in a certain proportion of cases, the latter disease not infrequently assuming a chronic course. The limits of acute simple nephrosis are widely apart. On the one hand is a mild degree of renal degenerative change accompanying a febrile disease and demonstrable clinically perhaps by albuminuria only; on the other hand, a disease very severe and associated with marked renal changes which may in part be responsible for the death of the patient. The latter changes are unusual under ordinary conditions of febrile or so-called toxic states, although known to accompany virulent infections and to follow the use of certain drugs.

It is to be expected that since acute simple nephrosis is generally a disturbance of short duration and frequently a terminal phenomenon of infectious and postoperative states it should occur in the presence of such long-standing renal conditions as arteriosclerotic renal disease, mild grades of hydronephrosis and local infections such as pyelitis. It seems reasonable to suppose not only that the nephrotic element of the picture is quite independent of, and therefore largely coincidental with, such other lesions, but that it is, in addition, not necessarily predisposed to by the preexisting disease. It is possible that with associated pyelitis, which is an inflammatory condition, a renal lesion should not be classified as nephrosis.

The occurrence of cases in which there is a combination of lesions must be recognized in order to avoid confusion concerning the etiologic interpretation of renal, and especially glomerular, changes. Since arteriosclerosis in particular is frequently associated with glomerular changes it must be distinctly recalled that in an arteriosclerotic kidney which is also the seat of nephrotic change the glomerular changes may more likely be due to the vascular than to the nephrotic condition. On the whole, the changes observed in cases in which there were combined lesions were not distinctive, and no unusual features seem to have been produced by a combination of diseases.

In addition, the changes in nephrosis are usually of short duration, since the disease processes from which nephrosis results are usually of short duration, leading either to death or to complete recovery. Consequently, acute nephrosis as a rule is not the "going concern" that chronic nephritis is, which, once on its course, usually goes to its completion.

The renal changes in acute simple nephrosis are generally described as follows: The kidneys are usually enlarged, smooth and pale or indistinct in color. The organs may appear boiled or yellowish. The edges of cut sections often become everted, and the sections are frequently opaque and present indistinct cortical markings. The surface may appear fatty. On microscopic study the glomeruli are usually reported to be normal, and in the tubules there usually are any number of changes from cloudy swelling and fatty degeneration to necrosis. Formation of casts is frequent. Finer studies of the glomeruli reveal occasionally: (1) swelling or various forms of degeneration of the epithelial cells of the tuft, the degree of degeneration frequently being similar to that present in the associated tubules; (2) desquamation of the epithelial cells of the tuft; (3) granules of albuminous material in the capsular spaces, variable in amount and occasionally appearing like hyalin; (4) more rarely, some swelling of the endothelial cells of the tuft or slight swelling of the basement membrane, and (5) collections of variable numbers of polymorphonuclear and mononuclear cells in the capillaries. Necrosis of the tufts has been reported but is very rarely seen under these conditions. Edema of the interstitial tissue may also be observed.

The present study deals with forty-one cases. In the majority of these cases the renal changes developed subsequent to operations on organs other than the kidneys. In other instances severe infections, such as septicemia, led to a fatal termination. In numerous instances obstruction of the gastro-intestinal tract was present.

The majority of the kidneys were grossly similar to those noted previously; that is, they were enlarged smooth kidneys, pale or indistinct in color, often appearing boiled or yellowish, and the cut sections presented opaque and frequently indistinct cortical markings, with edges that became everted. In those kidneys in which there was associated arteriosclerotic change or hydronephrosis the findings characteristic of these lesions were generally present also.

In the consideration of the microscopic observations of the glomeruli it was almost impossible to segregate them satisfactorily; consequently a more or less arbitrary gathering into divisions and subdivisions has been resorted to (table 1).

*Division 1. Cases in Which There Were Distinct Glomerular Changes.*—Ten cases fell into this division. In the majority (six cases) the changes could be ascribed to associated hypertensive or arteriosclerotic

change in the kidneys. In the minority (four cases) the explanation of the glomerular changes was less definite. In one of the four cases the patient was aged 59 years, and the renal changes may have been early senile changes. In another instance the kidneys were those of a boy of 13 years who succumbed to a very virulent infection, and the changes, as well as the clinical findings, were more suggestive of acute glomerular nephritis. In the remaining two cases the glomerular changes could not definitely be ascribed to any particular process.

TABLE 1.—*Arbitrary Division, for Purposes of Study Only, of the Forty-One Cases of Acute Simple Nephrosis*

Division 1	Division 2	
	Cases in which glomerular changes were distinct (10 cases)	Cases in which glomerular changes were not distinct (31 cases)
Cases in which glomerular changes were distinct (10 cases)	Subdivision 1 Cases in which a clinical diagnosis of renal disease, insufficiency or uremia had been made (11 cases)	Subdivision 2 Cases in which a clinical diagnosis of renal disease, insufficiency or uremia had not been made (20 cases)

*Division 2. Cases in Which There Were No Distinct Glomerular Lesions.*—Thirty-one cases fell into this division. For purposes of study they were divided into two subdivisions composed, respectively, of eleven cases in which a clinical diagnosis of associated renal disease, insufficiency or uremia had been made and twenty cases in which such a diagnosis had not been made. It was thought that such subdivision might lead to observations that would in turn elucidate the problem of the correlation of anatomic and physiologic disturbances, for in kidneys from patients who gave clinical evidence of renal disease such observations would be of distinct value. In these cases microscopic study showed the glomeruli to be essentially normal. The majority of them presented normal amounts of blood within the capillary loops and tufts of approximately normal size.

However, in many of the glomeruli minor changes were to be noted. They are considered minor from the anatomic point of view, although from the physiologic point of view it is conceivable that they may have been of great significance. These changes included: (1) granular débris, generally considered to be disintegrated cytoplasm or possibly coagulated albumin; (2) swelling, degeneration and desquamation of the epithelial cells of the tufts; (3) occasionally irregular thickening of the glomerular basement membrane, and (4) swelling or at times even slight increase in the number of the endothelial cells within the tufts. Such changes can readily be ascribed to degenerative processes and are not infrequently seen in the kidneys of patients who have died of acute infections, most of whose parenchymatous organs are affected by cloudy swelling.

The tubular changes were chiefly cloudy swelling and degeneration of the cells, producing partial obstruction of the tubular lumens; frequently there was dilatation of the tubules, at times very marked and associated with narrowing of the cell layer, together with degeneration of various types and extent. Granular débris in the tubular lumens was present in every case, whereas hyaline casts were less frequently noted. In addition to the usual changes noted in these kidneys, in three cases there was evidence of marked interstitial edema that not only separated the tubules from each other but also separated the intertubular capillaries from the tubules.

Another type of acute simple nephrosis is that encountered in cases of hyperemesis gravidarum. A consideration of the changes observed in such cases has been made so recently by Bell<sup>4</sup> that detailed consideration of the changes will not be made in this paper. The essential change is tubular degeneration. The glomeruli generally appear normal.

*Contrast Between the Two Subdivisions of Cases of Division 2.*—The clinical findings in the eleven cases included in the first subdivision were definite. The clinical diagnosis of renal disease, insufficiency or uremia was made because of elevated values of blood urea, significant urinary findings and occasionally edema, hypertension and oliguria. In every instance except one, abnormal urine was noted during the course of the disease; there were relatively large amounts of albumin, numerous casts and occasional erythrocytes and pus cells in the urinary sediment. In three instances sugar was also present in the urine, but this may perhaps be accounted for by the fact that solutions of dextrose had been administered intravenously. In many instances the specific gravity of the urine was 1.020 or more, and the urinary output in many of them, fair or good (from 500 to 1,000 cc. daily). Urine with this degree of specific gravity and in this daily volume is not generally found in ordinary cases of renal insufficiency, regardless of the cause. It must be noted that the relatively high specific gravity in some of these cases may have been due to the large amounts of albumin in the urine. Edema was absent in all except two of the cases. A study of the values of the blood urea in all cases except one was made, and in each instance the value of the urea was definitely above the normal; the highest value recorded was 388 mg. in 100 cc. of blood. It is interesting that in one case nephrectomy had been done several years previously for an independent condition.

In the second subdivision (twenty cases), which included those cases without clinical evidence of renal disease, the following features were noted: Studies of the urine on the days preceding death revealed a good output in ten cases, a fair output in seven cases, a poor output in one case,

4. Bell, E. T.: Am. J. Path. 8:1, 1932.

and no data in two cases on the days preceding death. In other words, oliguria was not a distinctive feature. In only four of the cases were the urinary findings distinctly abnormal; in seven cases there were slight abnormalities, such as a small amount of albumin or a few casts; in three cases the urine was normal, and in six cases the condition of the urine was not noted. The specific gravity of the urine in nine cases was 1.020 or higher, and was recorded as less than 1.015 in only three cases. Estimates of blood urea were made in ten cases; in seven of these the value was elevated terminally, but in one case only was the value more than 100 mg. in 100 cc. of blood.

From the clinical standpoint, therefore, the difference between the two groups was one of degree only, for qualitatively the changes were similar.

Pathologically, in contrasting kidneys of these two subdivisions, several variations were noted. The differences were not qualitative but quantitative and, as is to be expected, in the first subdivision (cases in which clinical diagnoses of renal disease, insufficiency or uremia had been made) the findings clinically and anatomically were more prominent than in the second subdivision, cases in which clinical diagnoses had not been made. The renal changes noted in this subdivision consisted essentially in the greater prominence of the minor changes already noted as being found in the glomeruli in cases of these two subdivisions. The more outstanding renal changes were, most likely, dependent on the more profound "toxemia" which had resulted from the serious disease that secondarily had led to renal injury.

*Case 1.*—No one of the cases can be considered typical of the entire group of cases of acute simple nephrosis. However, one of them is of such interest from the standpoint of demonstrating the failure of correlation between clinical and pathologic features in these cases that it will be reported in detail.

A woman, 45 years of age, came to the clinic July 5, 1928, because of loss of vision. The past medical history was essentially negative save that she had had numerous acute infectious diseases. The menopause had occurred at the age of 27 years, without apparent cause, and since that time there had been a gain of 75 pounds (34 Kg.). Approximately a year and a half before her visit dimness of vision gradually developed, and was more marked in the left eye than in the right.

Physical examination gave essentially negative results. The woman was obese, however. Blood pressure on two occasions was respectively 110 and 135 mm. of mercury, systolic, and 88 and 85 mm. diastolic. The urine had a specific gravity of 1.024 and 1.028, was acid in reaction, and contained albumin graded 3 and 4, an occasional hyaline cast and many granular casts. The return of phenolsulphonphthalein was 50 per cent in two hours. The value of the blood urea was 26 mg. in 100 cc. of whole blood, the value of protein in 100 cc. of serum was 6.1 Gm., and the fats totaled 525 mg. in 100 cc. of blood. The basal metabolic rate was —3.

A roentgenogram of the skull disclosed that the sella turcica was markedly enlarged, and that the clinoid processes were completely flattened. Ophthalmologic studies revealed a visual defect in each temporal quadrant.

There was no history of previous renal disease. In view of the absence of evidence of renal insufficiency and the good general condition of the patient, aspiration of a pituitary cyst was carried out July 18. The tumor proved to be adenocarcinoma, graded 2.

The postoperative condition was satisfactory for twenty-four hours, after which the blood pressure began to rise and reached a peak of 190 mm., systolic, and

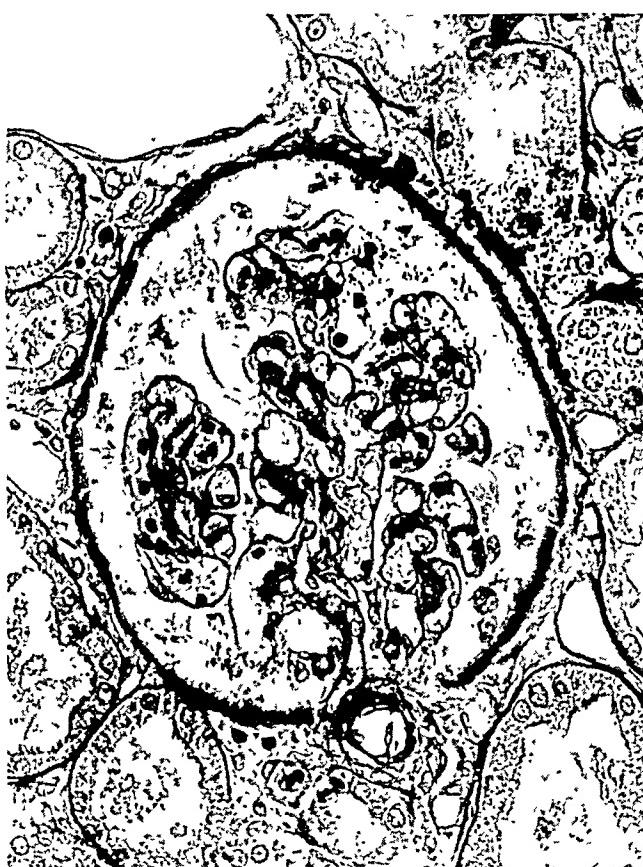


Fig. 1 (case 1).—Renal glomerulus from a patient with essential hypertension and simple nephrosis as a terminal event. The basement membrane of the tuft is thickened, particularly in the left half of the photomicrograph ( $\times 350$ ).

92 mm., diastolic, following which it steadily dropped during the twelve hours preceding death. The urinary output gradually decreased; the day following operation it was 100 cc., and the following day, 200 cc. Urinalysis disclosed albumin graded 4, a few hyaline casts, and occasional erythrocytes in the urinary residuum. The value of the blood urea rose to 139 mg. in 100 cc. The intake of fluid during the forty-eight hours between the operation and the time of death was 4,200 cc. Visible edema did not occur. Death occurred July 21.

The primary and secondary causes of death were recorded as carcinoma of the pituitary body and chronic diffuse nephritis (nephrosis). The right kidney weighed 169 Gm., and the left, 160 Gm. The capsules stripped with slight diffi-

culty, leaving smooth, purplish-red surfaces which were opaque in appearance. The glomeruli appeared normal in size and number, and presented the usual number of erythrocytes. There was considerable débris in the capsular spaces. The epithelial cells of the tufts were possibly slightly increased in number. Many of them were large and swollen. The endothelial cells appeared to be normal. The glomerular basement membrane appeared normal, although it presented occasional areas of thickening in the hilar regions of the tufts. The blood vessels and interstitial tissue appeared normal, while the tubules disclosed some dilatation and degeneration, with areas of thinning of the cellular cytoplasm. These changes were marked in some areas; elsewhere, the tubules appeared normal, although they contained much granular débris and some hyaline casts (fig. 1).

This case is interesting. The clinical evidence of renal disease was limited to the urinary findings, which were marked and consistently found. There was no evidence of edema or of lowering of the value of the serum protein, so characteristic of lipoid nephrosis. The normal appearance of the glomeruli was surprising in view of the marked urinary changes, and is illustrative of the fact that albuminuria may occur in the absence of apparent anatomic glomerular changes.

#### COMMENT

Several interesting problems are raised in a consideration of the pathogenesis of the symptoms in cases of acute simple nephrosis. The renal lesions are generally considered to be secondary to a primary disease, to arise as a result of so-called toxemia, and to be degenerative. As one might expect, the most highly differentiated tissue in the kidney, that is, the cells of the tubules, is the one most markedly affected. However, the glomeruli may participate to the extent of demonstrating mild degenerative changes, often proportional to the severity of the primary process. Consideration of all of the many clinical features observed in cases of acute simple nephrosis is beyond the scope of this presentation. However, it will be of interest to consider certain features which have a significant bearing on the subject.

The changes in the urine are those which would be anticipated: albuminuria, cylindruria and the occasional presence of erythrocytes and leukocytes in the sediment. From causes which are frequently, in part, extrarenal the urinary volume may be low, and marked oliguria is not infrequent. This process has been called the result of prerenal deviation of fluid or the retention of water by the tissues, so that a sufficient amount is not present for adequate formation of urine.

In considering some of the clinical features of cases of nephrosis, especially those associated with intestinal obstruction and conditions of dehydration, much discussion has been given to the mechanisms resulting in alterations in the chemical constituents of the blood, with particular reference to elevation of its nitrogenous content. Since the renal changes in these conditions are secondary to disease elsewhere it frequently has

been pointed out that the chemical changes which occur in the urine, and particularly in the blood, are not of necessity due to, or explained by, the renal changes only. Controversy has arisen, therefore, as to whether or not the elevation of the values of the blood urea in some of these cases is the result of renal or of so-called extrarenal factors, or of a combination of the two. Included in the extrarenal factors may be such mechanisms as dehydration, increased production of urea resulting from increased destruction of protein, and such alterations in the chemistry of the body as lead to retention of urea for preservation of osmotic relationships and so forth. The part played by the injured kidney cannot always be estimated. As Peters and Van Slyke<sup>5</sup> stated: "That the efficiency of the kidney is diminished cannot be denied. That it is the chief cause of the azotemia is open to question." If renal injury is a factor in the elevation of the value of the blood urea in some of these cases, evidence as yet is not available to determine whether or not the renal mechanism retaining the urea is similar to that in cases of glomerular nephritis with uremia.

In the opinion of some investigators the principal cause of the symptoms of renal insufficiency in these cases is the oliguria with the result that insufficient quantities of urine are formed to allow excretion of the necessary waste products in adequate amounts. However, as indicated in some of the cases in the present series in which the volume of urine seemed adequate and the urine of relatively high specific gravity, this explanation apparently does not suit all cases.

Further evidence that will help to settle some of these problems must come from accurate studies made clinically during the course of acute simple nephrosis of the type described, with evaluation of the metabolism of water, urea, minerals and other waste products.

In many of these cases, gastro-intestinal retention was present secondary to duodenal and pyloric obstruction, and in other cases, it was secondary to peritonitis and intestinal obstruction. As pointed out by Brown, Eusterman, Hartman and Rountree,<sup>6</sup> such conditions are frequently associated with "toxic nephritis," a syndrome presenting characteristic chemical changes in the blood and urine, and presenting at postmortem examination renal lesions characterized by tubular epithelial degeneration of a fatty and granular nature, occasionally with deposits of calcium, and without marked changes in the glomeruli. Zeman, Friedman and Mann<sup>7</sup> studied similar cases and characterized the renal

5. Peters, J. P., and Van Slyke, D. D.: Quantitative Clinical Chemistry, Baltimore, Williams & Wilkins Company, 1931, vol. 1, p. 298.

6. Brown, G. E.; Eusterman, G. B.; Hartman, H. R., and Rountree, L. G.: Arch. Int. Med. **32**:425, 1923.

7. Zeman, F. D.; Friedman, William, and Mann, L. T.: Proc. New York Path. Soc. **24**:41, 1924.

lesions as entirely degenerative. They expressed the belief that the deposition of calcium in the cells of the tubules is not secondary to the administration of calcium salts. Changes similar to these have been found in many of the kidneys studied in this series. The absence of distinctive glomerular changes is confirmed in the majority of these cases, although elevation of the value of the blood urea was a prominent feature in many of them.

## II. BILE NEPHROSIS

In the presence of obstructive jaundice the kidneys as well as other organs are bile-stained. With the staining there occur toxic effects which are known as bile poisoning, bile toxemia and so forth. The mechanism producing this change has never been fully understood, nor has it been expressed better than by the vague term "toxemia" or "toxic effect"; nevertheless, considerable clinical interest and importance are attached to the effect produced by obstructive jaundice on organs other than the liver. The internist has appreciated the fact that relief of obstructive jaundice must not be too long delayed, because of its damaging effect on the liver and other organs, and the surgeon has discovered that the three great dangers in surgical treatment of obstructive jaundice are hemorrhage, renal insufficiency, uremia and hepatic insufficiency. Renal insufficiency may be precipitated in cases of obstructive jaundice by the shock incident to surgical operation or anesthesia.

Considerable speculation has arisen concerning the active agent that produces renal injury and insufficiency in cases of obstructive jaundice. As possible agents, bile salts, bile pigments, associated infections and vasopressor and depressor or other toxic substances liberated from the liver have been suggested.

Clinical, and particularly experimental, evidence has been presented to prove or to disprove the importance of these various factors in the secondary effects of obstructive jaundice. As evidence accumulates it seems to point more and more to the importance of certain toxic or pressor substances elaborated by the hepatic tissue, or perhaps resulting from destruction of the hepatic cells, rather than to bile salts, bile pigments or infections.

Evidence of an adverse effect on the kidney in cases of jaundice is demonstrated clinically by the occurrence of abnormal substances in the urine, the result of "renal irritation." This may be mild or severe, more likely the former. Apparently in the early stages of jaundice renal injury may be more marked, since albuminuria is then more prominent than it is subsequently if jaundice continues for some time. Fitz-Hugh<sup>8</sup> pointed out that in twenty-five consecutive cases of catarrhal jaundice in persons who were, for the most part, healthy young adults,

8. Fitz-Hugh, Thomas, Jr.: M. Clin. North America 12:1101, 1929.

twelve of the patients had a cloud of albumin in the urine, eleven had a faint to heavy trace, and only two had less than a faint trace during the early stages of the jaundice. Similar observations have often been made in cases of obstructive jaundice. Still more remarkable evidence of the relationship of hepatic and renal disease is presented in a case reported by Helwig and Orr,<sup>9</sup> in which traumatic pulpefaction of the liver without rupture and with extreme jaundice and diffuse hemorrhages into serous cavities led to oliguria, elevated values of the nitrogen and creatinine of the blood and extensive histologic changes in the renal tubules. A similar case had been reported by Furtwaengler<sup>10</sup> in 1927. In the case reported by Helwig and Orr, erythrocytes and albumin were found continuously in the urine during the six days of the illness, and suppression of the urine was present to the extent that not more than 200 cc. of urine was passed during any one day. In addition, the blood pressure rose to 150 mm. of mercury, systolic, and 80, diastolic. Generalized edema developed, and jaundice occurred on the second day after the accident. The level of the nonprotein nitrogen rose steadily from 75 to 250 mg. and that of creatinine from 3.7 to 25 mg. in 100 cc. of blood. The kidneys were very large, swollen and soft and together weighed 580 Gm. Microscopically, they presented marked degeneration and even necrosis of some of the tubular cells, chiefly those of the convoluted tubules. The epithelium of the more highly differentiated cells was generally pigmented with a fine dust of greenish granules. The glomeruli presented swelling of the epithelium lining Bowman's capsule, and in the space between the tuft and the capsule erythrocytes were scattered about. In the capillary loops the endothelium was somewhat swollen, and in the tortuous capillary channels a few polymorphonuclear leukocytes were seen here and there. Stains for fat revealed no evidence of fatty degeneration or embolism in the kidneys.

This report is given in detail because it presents in a very striking way the results of an "acute experiment" illustrating the profound effect of hepatic necrosis and jaundice on the kidney. Although, in view of the extensive hepatic injury, the whole picture may not properly be said to be due to the jaundice alone, nevertheless there is a similarity in this case to cases of marked and prolonged obstructive jaundice in that in such cases extensive and irreparable hepatic injury occurs also.

An apparent adverse effect on the liver of disease of the renal and urinary tracts has been described by Dourmashkin<sup>11</sup> and by Fitz-Hugh. They reported cases of cholemia apparently induced by and "following instrumentation of patients having obstructive lesions of the urinary

9. Helwig, F. C., and Orr, T. G.: Arch. Surg. **24**:136, 1932.

10. Furtwaengler, A., quoted by Helwig and Orr.<sup>9</sup>

11. Dourmashkin, R. L.: J. A. M. A. **90**:908, 1928.

tract with coexisting hepatic cirrhosis." It is suggested that infection may play a significant part in this unusual clinical picture. Further evidence of the relationship of hepatic and renal disease has been presented recently by Helwig and Schutz,<sup>12</sup> who reported clinical and experimental observations on a hepatic-renal syndrome which they considered distinct from bile nephrosis. They expressed the belief that the syndrome was due to a potent toxin produced from injured hepatic tissue acting more or less specifically on the kidneys.

The changes in the kidney occurring in cases of cholemic nephrosis were described by Fahr<sup>13</sup> as follows: The kidneys are slightly enlarged and are irregularly or uniformly stained with bile. Microscopically, in Bowman's capsule are bile-stained rounded or granular masses of albumin. The capsular spaces may contain granules of bilirubin pigment or numerous desquamated epithelial cells or fragments, and occasionally the glomeruli reveal fine fat droplets. Similar bile-stained albuminous masses occur in the tubular lumens, and they may be so numerous as to form casts. Dilatation of the tubules, with flattening of the epithelium, may be present in such areas of cast formation. The epithelial cells may be stuffed with bile pigments and may show simple albuminous or vacuolar degeneration, occasionally necrosis. Exudative phenomena are absent. Kaufman<sup>14</sup> reported that the tubular epithelium undergoes fatty degeneration or necrosis and exfoliation, an injury caused by bile acids. Quincke<sup>15</sup> noted that the glomeruli remain practically unstained by bile.

The present study includes thirteen cases in which a diagnosis of bile nephrosis or its equivalent was made at necropsy. The methods of study and preparation of tissue were the same as those recorded previously.<sup>2</sup> Obstructive jaundice was present in each case, and the clinical data suggest a rather uniform sampling of the types of cases in which obstructive jaundice is present ordinarily seen clinically. The jaundice lasted from two days to two years and was constant or intermittent and of varied depth. Most of the patients were more than 45 years of age, as obstructive jaundice is rarely fatal before this age. In no instance was there any history of renal disease prior to the onset of the jaundice, and in some instances studies failed to reveal evidence of renal disease before the patients came to operation. In a few cases moderate hypertension had been present. The findings referable to the

12. Helwig, F. C., and Schutz, C. B.: *Surg., Gynec. & Obst.* **55**:570, 1932.

13. Fahr, T., in Henke, F., and Lubarsch, O.: *Handbuch der speziellen pathologischen Anatomie und Histologie*, Berlin, Julius Springer, 1925, vol. 6, pt. 1.

14. Kaufman, Eduard: *Pathology for Students and Practitioners*, Philadelphia, P. Blakiston's Sons & Company, 1929, vol. 2.

15. Quincke, H., in Nothnagel, H.: *Spezielle Pathologie und Therapie*, Vienna, Alfred Hölder, 1899, vol. 18, p. 63.

urinary tract consisted chiefly of albuminuria, occasional formation of casts, and occasional erythrocytes and leukocytes in the urinary sediment. In some of the surgical cases evidence of increased concentration of urea in the blood developed after operation, and in nine of the thirteen cases the value of the blood urea was elevated prior to the death of the patients. In three cases gastric retention and vomiting may have been a significant factor in producing the elevation of the blood urea. In the three cases in which it was noted the value of the blood chloride shortly before death was within normal limits. In the majority of the cases the records show that a fairly satisfactory urinary output was maintained, and even in some of the cases with elevated levels of blood urea, oliguria was not a feature. Renal insufficiency or uremia was given as a primary or a secondary cause of death in four of the thirteen cases. Most of the patients died from six to twelve days after operation. This includes four cases in which "renal insufficiency" was considered a factor.

Pathologic data show that in only two cases in the group were the kidneys of normal weight. In the remainder the kidneys were enlarged, the largest kidneys in the series weighing together 549 Gm. The majority of the kidneys were smooth, bile-stained and soft or flabby. In the majority of cases the glomeruli appeared normal microscopically. In almost all cases the glomerular epithelium appeared normal, although in a few there was some swelling or increase in the size of the epithelial cells of the tuft with at times excessive desquamation. In only six of the thirteen cases was there evidence of increase in the number of endothelial cells, and in the majority of these cases the increase was slight and associated with some swelling of the cells. The glomerular membrane appeared normal in most of the sections, although in some the membrane was irregularly thickened, particularly in the central or hilar portions, a finding probably associated with age. The occurrence of granular débris, probably disintegrated cellular cytoplasm, in the capsule of Bowman was not infrequent.

Tubular changes consisted chiefly of dilatation of the convoluted tubules with narrowing and degeneration of the epithelial cells, occasionally swelling with partial obstruction of the tubules, considerable granular débris and occasional casts within the tubular lumens, as well as bile pigment in variable amounts either within the cells or in the tubular or intertubular spaces. Changes in the interstitial tissue were slight and consisted of occasional patches of fibrous thickening or localized areas of edema; abnormal collections of cells were absent. In the kidneys of the older patients in the group sclerotic patches with hyalinized glomeruli and atrophic tubules were not uncommon. In figure 2 may be noted a renal glomerulus from a patient with bile nephrosis.

CASE 2.—A man, aged 43 years, came to the Mayo Clinic on Sept. 24, 1927, because of attacks of colic in the right upper quadrant of the abdomen of four years' duration and jaundice of three weeks' duration. The attacks of colic were characteristic of gallstone disease.

Physical examination gave essentially negative results, with exception of jaundice and obesity. The blood pressure in millimeters of mercury was 134 systolic and 94 diastolic. Urinalysis on two occasions gave negative results; the specific gravity was 1.018 and 1.020. Erythrocytes numbered 4,040,000 per cubic millimeter of blood. The value for bilirubin was 20.2 mg. in 100 cc. of serum.

On October 1, cholecystectomy and choledocholithotomy for subacute cholecystitis of grade 4, with cholelithiasis and choledocholithiasis, were performed. The postoperative course was stormy. Three days after operation the value of the urea was 143 mg. in 100 cc. of whole blood. It subsequently rose to 268 mg. and dropped



Fig. 2.—Bile nephrosis: normal glomerulus; coagulated plasma in many of the loops ( $\times 325$ ).

on the day of death to 250 mg. At the same time the value of the creatinine was 8 mg. in 100 cc. of whole blood, and that of the chlorides was 590 mg. in 100 cc. of plasma. Urinalysis revealed albumin graded 1+, a moderate number of hyaline casts and occasional erythrocytes and pus cells; the specific gravity was not recorded. The urinary output remained adequate, 2,645, 2,200 and 1,775 cc. on the three days preceding death. Edema did not develop. The blood pressure had not been recorded before operation. The cause of death was recorded clinically as obstructive jaundice and bronchopneumonia.

At necropsy, the right kidney weighed 250 Gm., and the left, 230 Gm. They were soft, flabby, swollen, smooth and brown. The glomeruli appeared normal. They were of the solid type. The epithelial and endothelial cells of the tufts appeared to be essentially normal, as did the basement membrane, with the exception of slight patchy thickening. The tubules were markedly dilated, and the cells revealed swelling and degeneration and contained bile pigment. The vascular walls were slightly thickened.

In this case, in which there was no evidence of preexisting renal disease, but in which jaundice was marked, evidence of renal insufficiency and injury developed after operation despite an apparently adequate output of urine. At necropsy, the kidneys presented, grossly and microscopically, the changes characteristic of bile nephrosis.

The changes noted in these kidneys may probably be considered largely degenerative and secondary to some toxic substance or substances associated with obstructive jaundice. With the exception of those cases in which previous hypertension was associated or occurred in older persons, the glomeruli in the majority of the kidneys in this series appeared essentially normal anatomically. The apparent slight increase in the endothelial cells in some of the tufts is difficult to explain and perhaps is not significant. It was not accompanied in every case by evidence of an infectious process elsewhere in the body. Clinical evidences of glomerular nephritis were lacking. Bile nephrosis is generally considered to be a degenerative disease, and glomerular lesions are considered to be absent in such cases. That glomerular injury had not occurred in a given case could not be definitely disproved. However, the presence of albumin in the urine and the occurrence of granular, probably albuminous material in the capsular spaces are very suggestive that such injury had occurred, and these findings were present in the majority of cases. The possibility that the albumin so excreted was abnormal, that is, not normal blood protein, is to be considered, since Andrews<sup>16</sup> and his associates suggested that the albumin in the urine in cases of nephritis may be abnormal, perhaps of hepatic origin. It is reasonable to assume that if abnormal protein exists in the blood plasma and the glomeruli are normal it will be excreted. Hayman and Bender<sup>17</sup> recently reported observations on the intravenous injection of plasma from three patients with nephritis who were excreting large amounts of protein. In no case did this cause albuminuria in healthy recipients.

The mechanism which produces increase of urea in the blood is not well understood. It is generally considered to be the result of either renal or extrarenal abnormalities. In cases of renal disease, the increased concentration of urea in the blood is generally associated with severe glomerular injury and disease, as is seen in cases of nephritis and nephrosclerosis, in which there is failure of excretion of urea in adequate amounts. In cases in which there is increased concentration of urea resulting from so-called extrarenal factors, the mechanism of the retention is uncertain. Many hypotheses have been advanced, but as yet none is entirely satisfactory. It is suggested by some investigators

16. Andrews, E., and Thomas, W. A.: J. A. M. A. **90**:539, 1928.

17. Hayman, J. M., Jr., and Bender, J. A.: Arch. Int. Med. **51**:447, 1933.

that elevation of the value of the blood urea due to extrarenal factors is a compensatory mechanism to maintain the osmotic pressure of the serum at normal levels in the presence of lowered values of the blood chlorides. In the three cases in the present series in which the values of the blood chlorides were determined shortly before death, those values were within normal limits. From the standpoint of the cases studied in the present series, it seems reasonable to believe that the increase in the urea in the blood in some of the cases of bile nephrosis in which it occurred was due, in part at least, to so-called extrarenal causes. The lack of microscopically apparent glomerular alterations is of significance in this connection, as is the fact that lesions of somewhat similar type, but more marked, may be found in the tubules in cases in which elevation of the blood urea is not shown. In the present series, significant anatomic differences between the cases with and those without increase in the blood urea were not noted.

As has been mentioned, the exact etiologic relationship between the obstructive jaundice and the renal lesion is not clear. Evidence points more and more to the importance of a nephrotoxic substance produced as a result of hepatic injury. The significance of the products of bile retention is uncertain since severe renal injury may occur in the presence of hepatic disease independent of jaundice, as was shown clinically and experimentally by Helwig and Schutz, but the lesions produced in the cases they described they considered were not strictly comparable to those of bile nephrosis. Interestingly enough, glomerular lesions of note were not reported in their studies.

It should be pointed out that by no means all patients with bile nephrosis succumb; as a matter of fact, the majority of patients who show this clinical change recover promptly and completely. The occurrence of a condition associated with increased concentration of urea in the blood may be a very significant contributing factor in bringing an illness to a fatal termination. There is no evidence, however, that the lesion produced in the kidney is irreparable.

### III. CHEMICAL NEPHROSIS

Chemical substances which are poisonous produce varying effects on the kidneys, depending particularly on the nature and the concentration of the poison. Since the proximal convoluted tubules are the most highly specialized cells in the kidney, they are generally the most readily and most markedly affected. In addition, they come in contact with such substances in relatively high concentration because their functions result in concentration of the material in the tubular lumens. It is interesting to note that certain chemical poisons have a selective action and consequently in moderate doses may affect only a localized portion in

the renal functional unit. Suzuki<sup>18</sup> showed, for example, that chromium harms chiefly the first portion of the proximal convoluted tubules in the experimental animal, uranium the third portion, and mercuric chloride and cantharidin the terminal, transitional piece. If the dose is sufficiently small, these regions only will be involved, but if larger doses are given, more of each tubule becomes affected and even the glomerulus may be diseased. Although the majority of such toxic substances affect principally the tubules for reasons already mentioned, certain poisons such as crotalins (rattlesnake venom) produce profound glomerular lesions. Pearce<sup>19</sup> showed this glomerular lesion in the kidney of the rabbit to be an acute exudative one. It does not lead, however, to subacute or chronic glomerular nephritis.

Clinically, mercuric chloride produces in most cases acute chemical nephrosis, and the picture it produces has become classic. Attention will therefore be paid to this syndrome. The clinical picture is usually one of vomiting, abdominal pain and diarrhea with oliguria or anuria, and whatever urine is passed is usually albuminous, light in color and low in specific gravity, and may contain numerous formed elements including casts and erythrocytes. Elevation of blood pressure may occur, but edema is unusual. Gradually uremia develops and death occurs, or after a period of uremia slow and apparently complete renal recovery results. The clinical picture is at times distinct from that seen in the so-called renal insufficiency of acute nephritis, and the lesions in the kidneys differ widely also. In cases of poisoning with mercuric chloride, the kidneys, which are smooth and soft, appear grossly enlarged. They generally appear grayish white and anemic, suggestive of fatty changes, but some investigators believe that after the eighth day the kidneys are red and congested.

The majority of studies reported in the literature indicate that the kidneys in cases of poisoning by mercuric chloride reveal tubular lesions primarily, and that these lesions include all forms of degeneration and in particular necrosis. Regeneration of epithelial cells occurs, if any cells remain from which regeneration can occur. Deposits of calcium salts in the necrotic cells of the tubules are characteristic of, but not exclusively limited to, poisoning with mercuric chloride.

The glomeruli generally, according to Fahr and Kaufman, are considered to be normal anatomically. Fishberg<sup>20</sup> mentioned that although the glomeruli, apart from congestion, usually appear normal, some swelling and less often foci of necrosis and desquamation of the capsular

18. Suzuki, T., quoted by Aschoff, Ludwig: *Lectures on Pathology*, New York, Paul B. Hoeber, Inc., 1924.

19. Pearce, R. M.: *J. Exper. Med.* **18**:149, 1913.

20. Fishberg, A. M.: *Hypertension and Nephritis*, ed. 2, Philadelphia, Lea & Febiger, 1931.

epithelium may be observed. Held<sup>21</sup> in one case described extensive glomerular changes, consisting of swelling and thickening of the capillary walls, proliferation and desquamation of the epithelial cells of the tuft, which were filled with hyaline droplets, and much exudation in the capsular space. Although such extensive changes are unusual in cases of mercuric chloride poisoning in man, they may occur in the experimental animal. Oliver and Smith<sup>22</sup> reported that in experimental nephritis of frogs, produced by mercuric chloride, the glomeruli were occasionally more injured than the tubules. These glomerular changes consisted of collections of a fibrin-like substance in Bowman's capsule, then formation of a hyaline thrombus within the capillaries, edema and necrosis of the tufts, and hemorrhage into the spaces.

Although "chronic" renal lesions are not produced as a rule by mercuric chloride, such lesions may be produced by other toxic substances, such as uranium, and the experimental work with this substance has led to observations which eventually may be of outstanding importance in a consideration of the pathogenesis not only of renal lesions but also of lesions in other organs. These observations are centered around the fact that in dogs a single dose of uranium of sufficient proportions may produce a "chronic" progressive renal lesion. This observation cannot as yet be applied to cases of nephritis in man, but the possibility of a single insult leading to a chronic progressive lesion presents the basis for a very alluring hypothesis that may eventually explain the pathogenesis of some renal lesions of man.

In the present series are reported three cases of acute chemical nephrosis. The first case followed poisoning by mercuric chloride; in the second case the etiologic factor is unknown, but the clinical picture and the pathologic studies indicated that acute chemical nephrosis existed, and the third was due to chloroform poisoning.

CASE 3.—This was a typical case of mercuric chloride poisoning affecting a young woman who died of acute renal insufficiency (table 2).

At necropsy the right kidney weighed 181 Gm., and the left, 237 Gm.; the kidneys were smooth and purplish red. The capsules stripped easily. On cut sections the edges became everted, and the tissue appeared pink and opaque, but with the markings fairly distinct. Microscopically, the glomeruli appeared normal in size and number. The capillaries were open, although perhaps less blood than usual was within them. There was considerable granular débris in the capsular spaces. The epithelial cells appeared normal, but many were desquamated. The endothelial cells appeared normal, although a few may have been swollen. The glomerular membrane appeared to be in good condition. The convoluted tubules revealed extensive degeneration and necrosis of the cells and complete disorganization of structure. Many of the tubules were dilated and filled with necrotic cells. There were many areas with regenerating tubules and cells with mitotic figures,

21. Held, A.: Ztschr. f. d. ges. Exper. Med. **61**:323, 1928.

22. Oliver, Jean, and Smith, Pearl: J. Exper. Med. **52**:181, 1930.

and the occurrence of more than one nucleus was not infrequent. Fatty change was extreme. There was marked edema of the interstitial tissue, with congestion of the vessels in many areas and occasional collections of a few leukocytes.

CASE 4.—In this case which was probably due to mercuric chloride poisoning there developed after operation for supposed carcinoma of the rectum a state of acute renal insufficiency. Because of persistent anemia decapsulation of the right kidney was performed (eleven days before death). The kidney was reported to be pale and somewhat enlarged. Immediately following operation it became very red. There was a slight temporary increase in the urinary output (table 2).

The right kidney weighed 225 Gm., the left, 245 Gm. The right kidney was pale red and smooth, and its capsule had been removed at operation. On cut sections the markings were indistinct, and the cortex measured 0.6 cm. in width,

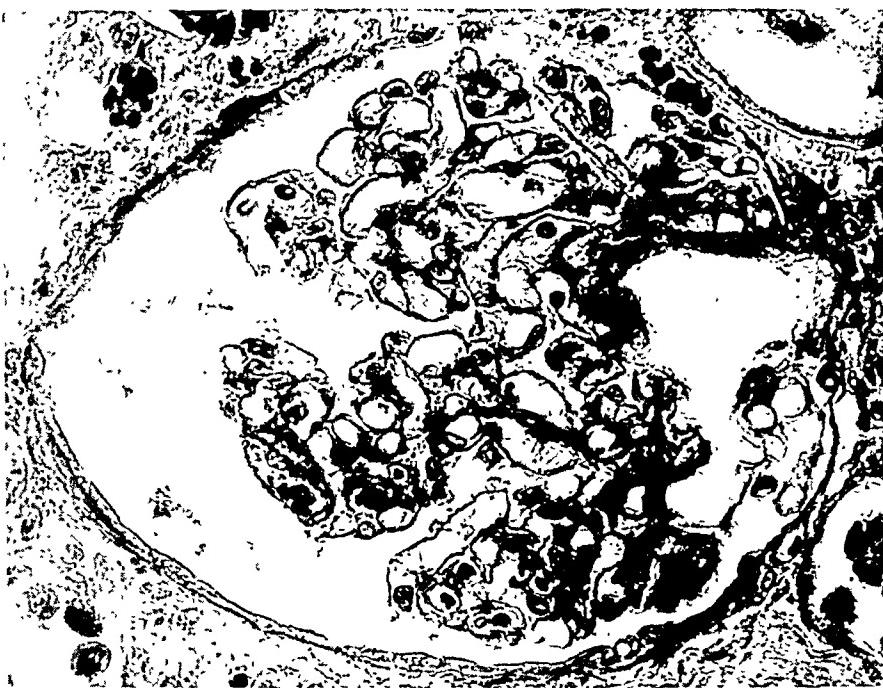
TABLE 2.—*Significant Clinical and Laboratory Data Noted in Several Cases of Nephrosis*

Case of Nephrosis	Cause of Nephrosis	Age, Years, and Sex	Blood Pressure, Min. of Mercury	Edema	Urine, Vol.	Urinalysis	Hemoglobin	Urea, Mg. in 100 Cc. of Blood	Creatinine, Mg. in 100 Cc. of Blood	Chloride, Mg. in 100 Cc. of Plasma	Cholesterol, Mg. in 100 Cc. of Plasma	Duration of Disease, Days
3	Mercuric chloride, 22.5 grains (0.16 Gm.)	23 F	132/68	++	150 cc.	.....	78%	288	13.2	462	114	7
4	Mercuric chloride (amount before poisoning not known)	56 M	140/76	..	Normal	Sp. gr. 1.036						
	After onset of nephrosis	150/30 190/40	Legs and back ±	75-275 cc. daily	1.010-1.012 albumin 1	28%	112 250 360	20.8	528	...	...	15 (?)
5	Chloroform 90 cc.	42 F	94/64 74/58	..	45-125 cc. daily	Albumin 2 Pus cells 3 Gm.	17.1 156 186	....	....	....	....	7
6	Eclampsia	19 F	160/90	+	.....	.....	..	...	...	...	...	2
7	Eclampsia	24 F	.....	0	0	.....	..	57	....	689	...	6 (?)

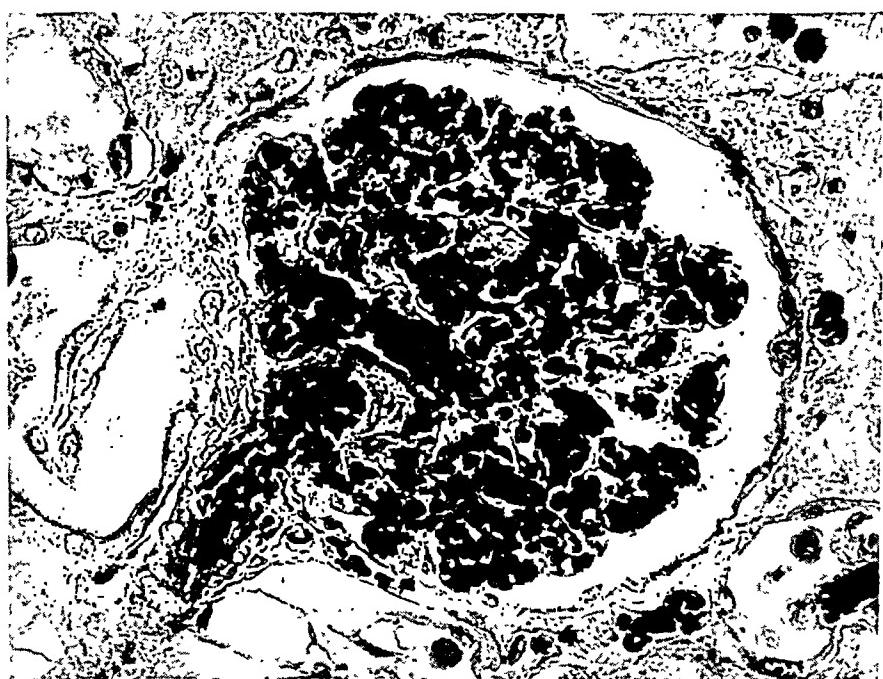
and the medulla, 1.6 cm. The capsule of the left kidney stripped easily and the kidney was light reddish brown; otherwise it was similar to the right.

The glomeruli appeared essentially normal microscopically. Approximately half of them were markedly congested. Some of these congested tufts were in groups, others were scattered, but most of them were nearer to the renal capsule than to the medulla. A few erythrocytes were in the capsular spaces. There was much desquamation of the epithelial cells. The parietal layer of Bowman's capsule contained cells which were somewhat swollen and prominent. The endothelial cells of the tuft were normal in appearance and number, although a few of them may have been swollen. The glomerular basement membrane was essentially normal. It revealed areas of thickening containing nuclei usually near the hilus of the tuft. A distinction between the two kidneys could not be made on the basis of their microscopic appearance.

The tubules were extensively degenerated. In many of them no cytoplasm remained, whereas in others remnants of cells were abundant. Much deeply staining material resembling calcium salts was present. Evidence of regeneration was



A



B

Fig. 3 (case 4).—Probable chemical nephrosis, but cause is unknown: *A*, normal tuft in left kidney ( $\times 400$ ); *B*, normal glomerulus with marked congestion ( $\times 400$ ).

almost lacking. A few tubules were filled with erythrocytes. Few casts were observed. Stains for lipoids revealed a few fat droplets in some of the cells of Henle's loop; occasional loose cells in the tubules were heavily laden with lipoid. The interstitial tissue was markedly edematous with possibly a slight increase in the amount of tissue. A few scattered lymphocytes were seen (fig. 3).

CASE 5 (reported in detail by Olson and Beaver<sup>23</sup>).—This case afforded a classic example of chloroform poisoning with suicidal intent. Jaundice (serum bilirubin, 10 mg. in 100 cc. with the direct van den Bergh reaction) and evidence of hepatic insufficiency in association with acute renal insufficiency were the outstanding clinical features (table 2).

At necropsy the right kidney weighed 152 Gm., and the left, 157 Gm. The kidneys were soft and mottled pink. The liver weighed 1,430 Gm. and was soft.



Fig. 4 (case 5).—Chemical nephrosis (chloroform): normal glomerulus ( $\times 325$ )..

On microscopic examination the glomeruli appeared essentially normal except for congestion. The tufts appeared of normal size, and there was very little débris in the capsular spaces. The epithelial cells of the tufts gave evidence of swelling and granular degeneration. The endothelial cells and glomerular basement membrane appeared normal. The tubules revealed varied amounts of degenerative change, with much lipoid in the cells. In some parts the tubules were dilated. In others there was much cellular disintegration, granular in type, although the nuclei were fairly well preserved. Casts were not present. The interstitial tissue revealed occasional regions of slight edema (fig. 4).

Evidences of serious glomerular disease were largely absent in these three cases, although the presence of débris, perhaps coagulated protein

23. Olson, P. F., and Beaver, D. C.: Death from Delayed Chloroform Poisoning, to be published.

in the capsular spaces, the occasional swelling and desquamation of the epithelium, and the glomerular congestion in the second and third cases suggested glomerular aberrations. But the changes were insignificant compared with the tubular alterations. There was no evidence microscopically in case 3 that the decapsulation had produced any permanent anatomic effect on the glomeruli.

It is interesting to speculate with regard to the pathologic physiology of the kidney in cases of poisoning with mercuric chloride. The anuria has been thought by some to be due to mechanical blocking of the tubules and to changes in the capsular epithelium. However, in view of the anatomic findings of blood in the capillary loops, the majority of which are open, it seems much more likely that the explanation offered by Richards<sup>24</sup> is correct. He demonstrated that in the kidney of a frog in which poisoning with mercuric chloride has occurred the glomeruli are extraordinarily active and glomerular fluid is separated at a faster rate than normal, and that this glomerular fluid is apparently of normal quality. Yet urine does not come from the ureter. In Richards' words: "The only explanation which I can reach is that under these abnormal conditions the osmotic pressure of the blood proteins is unobstructed by the normal qualities of the tubular epithelium and is able to draw all or nearly all of the glomerular filtrate back into the blood stream."

Certainly in two of the cases presented here the anatomic observations were compatible with such a hypothesis.

Few studies have been reported of the effects of chloroform poisoning on the kidneys. It is recognized that alteration in the lipoid content of the tubule cells, often manifested as fatty degeneration, is characteristic but not distinctive of this type of intoxication. Clinically, it is well known that patients poisoned with chloroform may demonstrate evidence of renal insufficiency. Whipple and Sperry,<sup>25</sup> and Williamson and Mann<sup>26</sup> have indicated that in experimentally produced chloroform poisoning the liver is the organ chiefly affected, but that other organs, including the kidney, may be injured.

In view of the profound degenerative changes in the liver produced by chloroform poisoning and the well known "toxic" effect on the kidney of the destruction of large amounts of hepatic tissue, as previously noted, it seems reasonable to speculate that in cases of chloroform poisoning a good deal of the apparent renal injury may be secondary to the associated destruction of hepatic tissue.

. 24. Richards, A. N.: Tr. A. Am. Physicians **44**:64, 1929.

25. Whipple, G. H., and Sperry, J. A.: Bull. Johns Hopkins Hosp. **20**:278, 1909.

26. Williamson, C. S., and Mann, F. C.: Am. J. Physiol. **65**:267, 1923.

## IV. RENAL CHANGES OF ECLAMPSIA AND PREGNANCY

The renal changes in eclampsia and other abnormal states of pregnancy have been so recently summarized and reviewed by Bell that at present nothing can be added, but confirmation of his results is possible in view of the findings in three cases studied. Bell divided his cases into five groups: (1) typical eclampsia with convulsions, (2) eclampsia without convulsions, (3) preeclampsia, (4) hyperemesis gravidarum and (5) pregnancy associated with preexisting renal disease. In the present study the renal changes in two cases of eclampsia with convulsions and in one case of hyperemesis gravidarum were observed.

The clinical appearance of eclampsia occurs late in pregnancy and is characterized by hypertension, occasionally by edema, albuminuria, oliguria, hematuria (microscopic) and convulsions. Retention of water and chlorides is common, but increase in blood urea is not common, and if present, may be due to renal or so-called extrarenal causes.

Bell, who studied the kidneys in fourteen cases of this type, using Mallory's aniline blue and Heidenhain's azan carmine stain, was able to confirm and extend the findings of Löhlein<sup>27</sup> and of Fahr. Grossly, the kidneys are usually slightly enlarged and their surfaces are smooth. The tissue in cut sections is usually cloudy and pale, although occasionally slightly yellow. Microscopically, the tubules present cloudy swelling or various forms of degeneration and even necrosis. The glomeruli are moderately increased in size, nuclear increase is variable but usually absent, and there is marked thickening of the glomerular membrane, with narrowing of the capillary lumens. This narrowing is due to massive thickening of the capillary basement membrane, which is not homogeneous but composed of parallel layers. Increase in number of the endothelial cells may be slight or striking, and the epithelial cells are only slightly altered, occasionally revealing fine droplets of fat or hyaline granules, but no proliferation. Bell differentiated the glomerular changes in eclampsia from those in clinical acute glomerulonephritis because in the former the "glomeruli are smaller, the basement membrane is much thicker, there are no polymorphonuclear leucocytes, no intracapillary fibers and no epithelial crescents." In glomerulonephritis he also found much more cytoplasm about the endothelial nuclei.

Bell interpreted the increase in number of the endothelial cells as an inflammatory phenomenon, that is, as a form of glomerular nephritis. The thickening of the glomerular membrane, he believed, is more difficult to explain; it may be an inflammatory or a degenerative lesion, the interpretation depending largely on one's definition of inflammation. He classified the lesion as a special form of glomerular nephritis. Others have classified it as nephrosis, considering it to be a degenerative lesion.

27. Löhlein, M., quoted by Bell. E. T.: Am. J. Path. 5:587, 1929.

and it has also been classified as a separate group, distinct from glomerular nephritis and nephrosis.

Following is a report of two cases of eclampsia:

CASE 6.—The clinical features in this case were characteristic of eclampsia, with convulsions, blindness and anemia (table 2).

At necropsy the right kidney weighed 155 Gm., and the left, 132 Gm. The capsules stripped with ease and the kidneys were smooth, moderately deep red, and on cut sections deep pink. The cortices were thickened and swollen, and the markings indistinct.

On microscopic examination the glomeruli appeared somewhat lobulated, enlarged and numerous. A moderate number of glomerular loops were open, but erythrocytes were rather scarce. In the capsular spaces a small amount of granular material was present, but crescents were absent. The epithelial cells of the tufts were swollen; some of the cells were very large, granular and pedunculated. Desquamation was frequent. The endothelial cells were increased in number and swollen in many loops; some of the loops were almost filled with several large endothelial cells. This process was not uniformly present. The glomerular membrane was uniformly thickened in most of the tufts, although in a few it revealed only slight, if any, thickening. The convoluted tubules showed moderate cloudy swelling and degeneration. In many areas the tubules were dilated, and granular débris and hyaline casts were present in moderate numbers. The interstitial tissue and vessels appeared normal. Stains for fat revealed scattered tubules containing fat droplets and an occasional glomerulus in which there was a deposit of fat in the endothelial cells of the tuft.

CASE 7.—The clinical features were as characteristic as those in case 6 (table 2).

At necropsy the right kidney weighed 170 Gm. The weight of the left kidney was not recorded. The capsules stripped easily from the renal surfaces, which were smooth and brownish red. On cut sections the markings were exaggerated, although they were somewhat indistinct near the periphery of the cortices. The cortices were 0.6 cm. wide, and the medullas were 1.5 cm. wide. The only other significant finding was acute degeneration of the liver.

Microscopically, the glomeruli appeared enlarged, many of them filling the capsular spaces completely. Considerable granular deposit was present in the spaces. The tufts were relatively bloodless, and the individual loops did not seem so wide open as they usually are in normal glomeruli. The epithelial cells of the tufts were swollen and some were desquamated. In most loops the endothelial cells appeared normal, whereas in others there was evidence of proliferation, with even complete obstruction of the loops by huge endothelial cells with abundant cytoplasm. A few intracapillary hyaline fibers were noted. The glomerular membrane was almost uniformly thickened. The thickening was fibrillar. A few loops revealed a membrane of normal thickness. The tubules appeared variable; some were dilated and had flattened, degenerating epithelium; others presented swollen cells, with much granular deposit in the lumens. Deposits of fat occurred in some of the cells of the proximal convoluted tubules. The interstitial tissue appeared normal, as did the vessels (fig. 5).

In the two cases of eclampsia studied, the renal findings were similar to those described by Bell. In both cases fibrillar thickening of the membrane was the outstanding feature in the glomeruli. The

glomeruli were relatively enlarged and relatively bloodless. They practically filled the capsular spaces. In both cases some loops revealed complete obstruction by proliferated endothelial cells, although this feature was not pronounced.

The renal lesion in eclampsia is probably a secondary one, and the kidney is only one of the organs affected. The origin of eclampsia is unknown. The condition is presumed by many to be a form of toxemia, the origin of which may be in the fetus. Another possible source of the toxic substance is the liver. It is known that a diseased liver produces a substance or substances which may lead to degenerative changes in the kidney. The details have been considered in the comment on bile



Fig. 5 (case 6).—Glomerulus in kidney of patient with eclampsia: obstruction of loops; relatively large tuft; thickened basement membrane; increase in size and number of endothelial cells of tuft ( $\times 430$ ).

nephrosis. However, the substance generally produced under such circumstances affects the tubules in cases of bile nephrosis, leaving the glomeruli relatively normal, whereas the renal lesions of eclampsia are primarily glomerular.

It is impossible to state the nature of the change in the glomeruli in the present study, but, as Bell pointed out, it is a distinct lesion whether one classifies it as degenerative or inflammatory; since there is a strong possibility that the lesion is purely vascular, secondary to hypertensive and spastic changes, it may not be inflammatory in origin. Perhaps the most interesting speculation regarding the lesion is its probable course following relief of the eclamptic symptoms. The question which

naturally arises is this: If the glomerular membrane is once thickened, does it ever become normal again, or does it remain thickened? As yet, postmortem studies of kidneys from women who have had eclampsia years previously are not recorded in the literature, so this question must for the present remain unanswered. Some data are available which indirectly suggest that when once a change occurs in the glomerular loops and membrane in cases of eclampsia, this change may be permanent. In studying the arterioles of the retina in cases of toxemia of pregnancy, Wagener<sup>28</sup> came to the following conclusions: In early cases with angiospastic changes only, complete recovery without subsequent evidence of vascular disease may occur. However, if organic changes in the vessels and diffuse retinitis develop as a result of more continued or more severe angiospasm, generally persistent hypertensive vascular disease will develop subsequently. It is known that, so far as function is concerned, the kidneys of a previously eclamptic woman return to normal, but experience has shown that moderate anatomic abnormality of the kidney may not always lead to evident functional change. Consequently this occurrence does not answer the question. When it is answered, physicians will know more concerning the nature and significance of the thickening of the glomerular membrane not only in cases of eclampsia but possibly also in cases in which the patients recover from clinical acute glomerular nephritis. It is also to be recalled that among those women with eclampsia who recover, the presence of thickening of the glomerular membrane is merely speculative and it may be that the change is very slight.

The glomerular lesion in eclampsia is so distinct in the sense that it is entirely different from that noted in any of the other forms of nephrosis that this lesion should probably not be classified pathologically as nephrosis. Its exact classification will depend on future observations. However, the renal lesion observed in cases of hyperemesis gravidarum is so similar to that seen in the different forms of nephrosis that it may be so classified at present.

Studies of the kidneys of a patient with hyperemesis gravidarum have been reported previously in this paper. The glomeruli in these kidneys appeared normal, although there was some granular débris in the capsular spaces.

#### SUMMARY

In this presentation I have described histologic studies of the glomeruli in the kidneys in cases of simple nephrosis in which the diagnosis was made after postmortem study. Cases of simple nephrosis were grouped as acute simple nephrosis, bile nephrosis, chemical nephrosis and renal changes of eclampsia and pregnancy.

28. Wagener, H. P.: Proc. Staff Meet., Mayo Clin. 8:461, 1933.

A group of ten cases of acute simple nephrosis presented distinct glomerular changes consisting primarily of irregular thickening of the glomerular basement membrane and less often of increase in number and swelling of the endothelial and epithelial cells of the tufts. In the majority of cases these changes were considered independent lesions, the result of associated hypertension, arteriosclerosis or other complicating renal disease. They did not resemble the lesions seen in cases of clinical glomerular nephritis.

In a second group of thirty-one cases of acute simple nephrosis the glomeruli revealed, as a rule, normal tufts with occasional minor variations consisting of variable amounts of débris, usually granular, in the capsular spaces, swelling, degeneration, slight increase in number of the endothelial or epithelial cells of the tufts and occasionally slight, irregular thickening of the glomerular basement membrane. These changes were considered to be probably degenerative, and not suggestive of, or similar to, those observed in cases of glomerular nephritis or lipoid nephrosis.

In a series of thirteen cases of bile nephrosis in which the diagnosis was made at necropsy the majority occurred in persons who presented glomeruli that appeared normal histologically, although a moderate amount of granular material was often noted in the capsular spaces. In six of the thirteen cases slight swelling or proliferation of the endothelial cells of some of the tufts was observed. The epithelial cells of the basement membrane appeared essentially normal. These changes were slight and could not be correlated with the clinical findings, and consequently their significance is doubtful.

In three cases of chemical nephrosis the glomeruli appeared normal except for congestion, desquamation of many of the epithelial cells and granular material in the capsular spaces.

The glomerular changes observed in two cases of eclampsia were similar to those described by Bell; there were moderate increases in size of the tufts, absence of nuclear increase, or a variable degree of it, and marked, irregular fibrillar thickening of the glomerular membrane. In one case of hyperemesis gravidarum the glomeruli appeared normal, although granular material was noted in the capsular spaces. The renal lesion in eclampsia is probably secondary, and although it is distinct and probably degenerative, one is not able to state its nature at present.

# ACTIVE AND PASSIVE PLEAT FORMATION OF JOINT CARTILAGE

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The ends of most normal joints are covered with an almost perfectly smooth layer of hyaline cartilage. In later years some irregularities may appear as a result of proliferation and degeneration in the joint cartilage, viz., fibrillation, erosion and even denudation of the subchondral bone, but the unevenness produced in this way is inconsiderable, and the level of the old surface is more or less preserved. I shall consider in this article only the free joint surfaces, exclusive of the joint borders, where in more advanced cases of deforming arthritis large marginal exostoses may develop. More marked deformities of the joint surface may be produced by osteochondritis dissecans after the joint body becomes free or by intra-articular fracture. A part of the end of the joint, with its cartilage and the subchondral bone, may break off, and after reattachment a small steplike deformity may be found at the site of the fracture. Löw-Beer<sup>1</sup> made a histologic study of several intra-articular fractures, but in no case of his series could a real plication of the joint surface be seen. I recently saw an intra-articular fracture of the knee joint in a case of Paget's disease, in which, despite overlapping of the fragments of the joint cartilage, the deformity of the joint surface itself was not pronounced.

In the following pages I shall discuss a type of unevenness of the joint surface in which the folds are large enough to be seen grossly. These folds indicate the presence of some pathologic process which leads eventually to a decrease of mechanical resistance of the joint cartilage.

The process of folding can be either active or passive. Both forms are the result of a disproportion in the dimensions of the bony epiphysis and of its cartilaginous cover. This disproportion may be relative or absolute. It is relative when the total volume of the bony epiphysis becomes diminished by destructive or resorptive processes and the joint cartilage—which is not primarily involved in the process—preserves more or less its normal size. It follows the diminution of the epiphyseal bone with the formation of folds, the apexes of which are directed

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1. Löw-Beer, A.: *Virchows Arch. f. path. Anat.* **273**:191, 1929.

toward the center of the epiphysis. This passive folding of the joint cartilage is thus necessarily combined with a diminution of the volume of the epiphysis; at best, its surface may remain the same, but, as a rule, it will become smaller.

In the active form the disproportion between cartilage and epiphyseal bone arises from augmentation of the volume of the cartilage by extensive proliferative processes in the cartilaginous tissue. By these processes the cartilage becomes too large for the bony epiphysis and forms folds, the apexes of which, different from those already mentioned, are mainly directed toward the joint cavity. By this process of active pleat formation the surface of the entire epiphysis becomes enlarged, but there is practically no change in the volume of the bony epiphysis.

To illustrate the active and passive process of cartilaginous pleat formation, I shall describe briefly a few cases, most of which have been reported before in other connections.

I shall first describe the passive process, which, as has already been indicated, is due mainly to changes in the bony epiphysis, especially in the subchondral zone. Unevenness of the joint surface may also be caused by inflammatory processes in the joints, combined with an increase in intra-articular pressure, necrosis, with softening of the joint cartilage, and porosis of the subchondral bone. These factors may bring about herniation of the joint cartilage toward the bone marrow. This is seen frequently in cases of tuberculous and suppurative arthritis, but it also occurs in animals after injection into the joints of various chemical substances which lead to an exudative arthritis. All these pictures are similar and represent the simplest type of passive formation of cartilaginous pleats. Macroscopically, the formation is barely visible. The microscopic appearance is illustrated by figure 1,<sup>2a</sup> which shows the joint cartilage of a patient with chronic suppurative arthritis, with prolapse of old softened and necrotic joint cartilage toward the subchondral marrow spaces, which have been enlarged by osteoclasts and chondroclasts. Finally, after removal of the zone of preparatory calcification and of the subchondral occluding bony lamella, the marrow spaces may become so large that they are covered only by a thin layer of noncalcified cartilage. This may easily be displaced by the high intra-articular pressure toward the spongy bone, with which it becomes connected secondarily by dense fibrous marrow. In tuberculous joints, in which, as a rule, the undermining of the joint cartilage is more extensive, herniation of the joint cartilage is more frequently observed, but it does not lead to a more marked deformity of the joint surface, because the entire cartilage usually becomes resorbed or sequestrated sooner.

2. Freund, E.: (a) *Virchows Arch. f. path. Anat.* **284**:384, 1932; (b) **261**: 287, 1926; (c) **274**:1, 1929; (d) **277**:326, 1930.

Passive folding of joint cartilage is better seen in cases in which slow reorganization of the bony epiphysis takes place, especially if in the process of reorganization the resorption of bone exceeds the formation of new bone. This is the case in aseptic necrosis of the epiphyses, which does not lead to sequestration of the necrotic area (as in osteochondritis dissecans), but to restitution following resorption of the necrotic tissues.

#### REPORT OF CASES

CASE 1.—An unusual case of aseptic necrosis of both femoral heads was observed in a woman, 77 years of age.<sup>2b</sup> Macroscopically, both heads were flattened, and the joint cartilage showed folding at the apex, as though it were too



Fig. 1.—Herniation of joint cartilage toward the subchondral marrow spaces in a case of suppurative arthritis. The subchondral bony lamella and the calcified layer of joint cartilage show wide discontinuity; the defect is filled with fibrous marrow toward which the noncalcified cartilage is prolapsing; the joint cartilage is thinned out from above, and its superficial layers are impregnated with fibrinoid substance.

large for the subjacent spongy bone. In both heads the necrosis involved a more or less wedge-shaped area in the subchondral zone. A fracture line passed through the necrotic bone immediately below the joint cartilage and parallel to the joint surface. Although the fracture was old, it showed no signs of healing because it passed for its entire length through necrotic bone which was distant from vascularized living bone marrow.

The continuous friction of the fracture surfaces on each other leads to the accumulation of bone detritus in the fracture space, which gradually becomes massaged into the neighboring marrow spaces. The loss

of bone tissue just below the joint cartilage deprives the latter of its solid support.

However, not only the subchondral fracture, but also a demarcation zone at the borderline between necrotic and healthy bone, weakens the solid structure of the subchondral zone. Necrotic bony trabeculae



Fig. 2 (case 1).—Deep implication of the joint cartilage in aseptic necrosis of the femoral head. Surface of the cartilage is covered by a fibrous tissue pannus; at the apex of the cartilaginous fold two islands of newly formed cartilage are present in the bone marrow.

become gradually more and more resorbed. Moreover, the formation of new bone does not keep pace with the resorption of bone, and this disproportion leads to weakening of the firmness within the bony epiphysis and to consequences detrimental to the joint cartilage. Finally, after

removal of all the subchondral necrotic tissue, the demarcation zone with its fibrous bone marrow reaches the lower surface of the joint cartilage, and fibrous and cartilaginous tissues grow together. But such a change can be seen only in places with a relatively small extension of the necrosis, especially at its margin, where from the beginning the necrotic area is less extensive in its vertical diameter. In certain areas (fig. 2) the joint cartilage may show arcuate or undulate downward



Fig. 3.—Comminuted fracture of the joint cartilage, with displacement and bending of the fragments in same case as in figure 2. The subchondral marrow spaces are shown after resorption of necrotic bone and bone marrow, filled with fibrous tissue, which has also invaded the joint cartilage, bringing about its resorption from below.

displacement. The zone of preparatory calcification and the underlying occluding bony lamella, which are unable to expand because of their rigid consistency, show numerous radial fissures and fracture lines. These traumatic lesions form the first points of attack for the resorption of calcified and even noncalcified cartilage, which thus merges gradually into fibrous bone marrow.

The displacement of the cartilage may be so great in some places that the cartilaginous cover breaks into fragments of different size, forming an irregular heap of cartilaginous bodies (fig. 3). The fibrous bone marrow lies directly below the folded and broken cartilage, which gradually becomes resorbed. Large sinuses form in the deeper layers of the joint cartilage and are partially filled with fibrous tissue. In some places, however, the subchondral fibrous bone marrow shows transformation into cartilaginous tissue. This is especially true where the apex of the cartilaginous fold reaches the underlying fibrous bone marrow or where spongy bone is forming a prominence toward the subchondral fibrous tissue—in other words, in those places where motion of the joint brings about friction between hard substances (as bone tissue and calcified cartilage) and fibrous tissue. It is known that friction and gliding motion are predisposing factors in cartilage formation.

In other areas the cartilage is broken through entirely. The subchondral fibrous tissue grows toward the joint surface through these fractures and fissures in the joint cartilage, forming a fibrous pannus in and around the fracture line of the cartilage. This tends to reunite the fragments. Many small pieces of cartilage and bone are embedded in such a pannus. It is interesting that the pannus, by bridging the valley of the cartilaginous fold, apparently decreases the unevenness of the joint surface. Thus the joint surface may appear smooth, but unevenness exists beneath it.

CASE 2.—I observed essentially the same process of passive fold formation of the joint cartilage, but with fewer signs of trauma, in a woman, aged 71, suffering from Paget's disease (fig. 4).<sup>2e</sup> The case was complicated by fracture of the neck of the femur, and only the broken femoral head showed unevenness of its surface. The entire head was extremely porotic, and practically all of the old spongy bone and the old subchondral bony lamella had disappeared. The joint cartilage was greatly thinned out by resorption from its lower surface, where a layer of fibrous marrow had formed, with bone typical of Paget's disease. The joint cartilage had become deprived of its normal solid support, just as in the patient with aseptic necrosis, but to an even higher degree. Even a slight temporary increase in intra-articular pressure was too great for this bone to withstand, and the joint cartilage easily became displaced toward the marrow spaces in four deep parallel folds.

At the deepest portions of such implications the joint cartilage may again show complete interruption. At these points fibrous bone marrow can push its way toward the joint cavity and cover the joint surface as a pannus. It is of interest that Paget's bone forms in this fibrous tissue on the upper side of the joint cartilage, and so it is possible to find bone marrow typical of Paget's disease and trabeculae with mosaic structure within the joint cavity.

This joint deformity can by no means be called a typical sign of Paget's disease. Against such a supposition is the fact that in the

same case the other femoral head showed a perfectly smooth joint surface, although in its subchondral zone the changes of Paget's disease were more pronounced. The main predisposing factor in the development of such a severe degree of joint deformity is the marked osteoporosis of the epiphysis, which weakens the bony support of the joint cartilage. But the traumatic factor, represented in its simplest form by a temporary increase in intra-articular pressure, is indispensable. I have seen a great number of cases with the highest degree of bone atrophy, especially in the femoral head (after fracture of the neck of the femur), but in none of these was there any cartilaginous pleat formation. I have also seen cases of so-called osteomalacia carcinomatosa, in which most of the bones were almost completely replaced by tumor tissue and formed a mass of soft waxlike consistency. The



Fig. 4 (case 2).—Folding of the joint surface in a case of Paget's disease. The head of the femur shows extreme osteoporosis; the joint cartilage shows fracture lines through which the diseased bone marrow reaches the joint cavity.

patient was bedridden and thus was protected against any considerable trauma, which explains the perfect preservation of the shape of the bones and the joint surfaces.

CASE 3.—Probably the highest degree of deformation of the joint surface as a consequence of passive implication of the joint cartilage was shown in a woman, aged 78, suffering from Recklinghausen's osteitis fibrosa (figs. 5 and 6). The right femur showed extreme osteoporosis, with a great number of "brown tumors" in the bone marrow. Fractures were visible at several places, most of which had healed, with marked deformity, although some were of more recent date. An unusual picture was seen at the proximal end of the femur. No normal bone structure was left, and the few irregular and coarse bony trabeculae accompanied by strips of fibrous bone marrow were not able to support the joint cartilage. It was greatly thinned out and in a few places had disappeared. In such places, i. e., surrounding the fovea capitis femoris, the brown bone marrow was covered only

by a thin pannus of fibrous tissue. Owing to the decreased volume of the bony epiphysis, the joint cartilage showed waviness over all the joint surface, in some areas giving the impression that it had become reefed in a regular way, so that it resembled a gathered piece of cloth.

The cause of the fold formation in this case was, as may clearly be seen by the histologic picture, essentially the same as in the case of Paget's disease. There

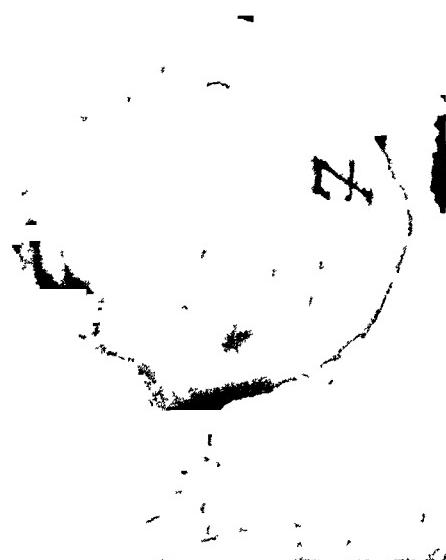


Fig. 5 (case 3).—Photograph of the head of the femur, showing irregular joint surface and folding of the joint cartilage.



Fig. 6.—Extensive folding of the joint cartilage and markedly porotic bone with "composed" bony trabeculae and fibrous bone marrow in the subchondral zone in the same case as in figure 5. Many of the valleys have become filled with new cartilaginous tissue.

was extreme osteoporosis in the femoral epiphysis, and the subchondral zone was occupied by not very resistant fibrous marrow, with, in greatest part, osteoid fibrous bone. But there were some differences. The displacement which took place in the joint cartilage was not merely passive as was almost entirely the case

in the patient with Paget's disease. There were many reactive changes in the joint cartilage, despite the patient's age. The main feature was the atrophy, which had brought about the marked thinning out of the cartilaginous cover. This could be seen already grossly.

Microscopically, one can follow this process of atrophy in its different stages. The first stage is enlargement of the cell capsules in the more superficial layers of the joint cartilage. The cartilaginous cells proliferate and assume the spindle shape of fibroblasts, or become starlike, and the surrounding hyaline ground substance disappears gradually (Weichselbaum's lacunae, fig. 7). The interlacunar septums become thinner and are finally melted down, and the lacunae merge. By this process large portions of the joint cartilage are lost and are replaced



Fig. 7.—Photomicrograph showing: disappearance of joint cartilage by resorption from above and below, confluence of Weichselbaum's lacunae and formation of a loose fibrous tissue pannus on the joint surface, resorption from below by fibrous bone marrow with trabeculae of fibrous bone and proliferative changes of the cells in the deeper layers of the joint cartilage.

in part by loose connective tissue. This becomes vascularized later, forming a connective tissue pannus on the joint surface of the femoral head.

#### COMMENT

It is of interest that the process of atrophy does not give evidence of catabolism only. Anabolic changes can take place, and the fibrous pannus can again lead to cartilage formation. Then one finds old hyaline cartilage in direct connection with young, extremely cellular cartilage of a more embryonic type. These changes take place especially in the valleys of the uneven joint surface. The production of cartilaginous tissue may be so marked here that the valleys again become completely

filled with cartilaginous tissue (fig. 8). This corrects the unevenness of the joint surface. In other places the waviness is equalized only by the connective tissue pannus, which runs straight over the cartilaginous folds, hiding them completely from above. This explains the difference in counting the folds on the gross specimen and later on the microscopic slide. Grossly, there were only five parallel folds, whereas, histologically, their number amounted in certain places to as many as ten.

If an old person is able to show so many reactive changes in the joint cartilage, one may, of course, expect more in a young person, in whom a combination of active and passive processes in the joint cartilage may yield even more marked deformities.

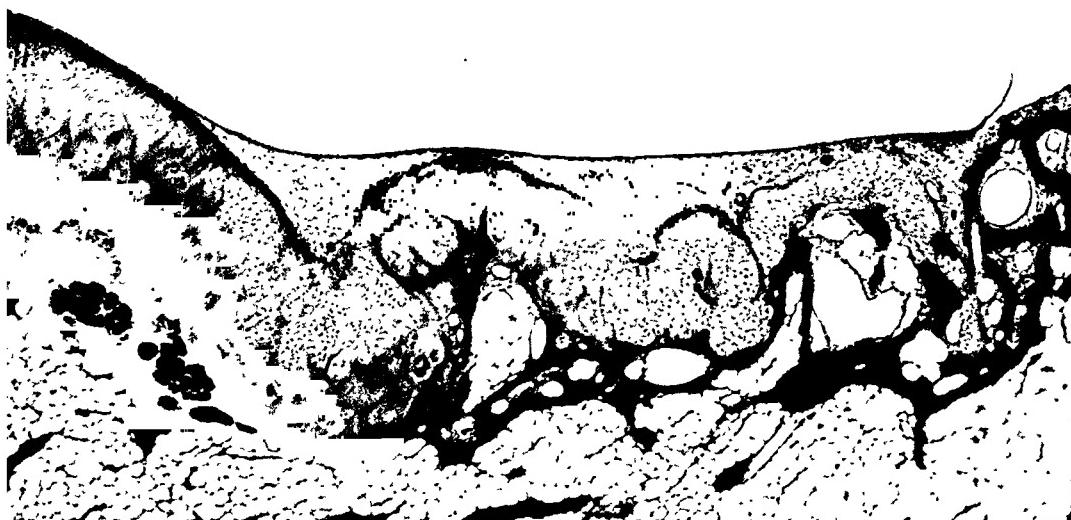


Fig. 8.—Correction of the unevenness of the joint surface by the formation of a fibrous pannus which covers the cartilaginous folds entirely. Active proliferation of the cells of the old joint cartilage followed disappearance of the hyaline ground substance.

In a case of Perthes' disease, for instance, in which the whole femoral epiphysis becomes necrotic and undergoes reorganization, merely passive cartilage implication may occur, especially under weight-bearing and during the active stage of the process. The disproportion in size between the bony nucleus and its cartilaginous cover increases, however, as the process goes on, and this occurs not only by resorption of the necrotic tissues but also by proliferation of the joint cartilage. It is well known that the joint cartilage remains alive in cases of aseptic necrosis of the subchondral bone, deriving its nutrition entirely from the synovial fluids. In a patient with Perthes' disease the joint cartilage is still growing and may be even larger than normal. In normal subjects there is a continuous resorption of cartilaginous tissue from the bone marrow,

owing to the process of enchondral ossification. In Perthes' disease, with its necrotic bone marrow, there is cessation of enchondral ossification of the joint cartilage during the whole period of reorganization. This must lead to an abnormal accumulation of cartilaginous tissue, if the growth of the joint cartilage goes on within normal limits, as it apparently does. The cartilaginous cap of the epiphysis must outgrow the epiphysis and must, therefore, adjust itself to this disproportion by the formation of folds. And, as a matter of fact, roentgenograms in cases of Perthes' disease show, especially in the so-called stage of fragmentation, irregularities of the joint surface indicated by steps and discontinuities in the subchondral bony lamella. The joint space is wider than on the normal side, which is a consequence of the increase in thickness of the joint cartilage. From these signs in the roentgenograms and especially from descriptions in textbooks, one may conclude that deformities of the joint surface, with folding of the joint cartilage, exist. In Perthes' disease they are due to an active and passive deforming process of the end of the joint and especially of the joint cartilage.

This mixed form leads to a discussion of that group of implications of the joint surface which is almost exclusively the result of active proliferative changes in the joint cartilage itself. The changes in the subchondral zone are of little or no importance. I studied this group in its purest form in cases of tabetic arthropathy.

Moritz<sup>3</sup> has shown in his excellent study that reestablishment of the enchondral ossification of the joint cartilage is characteristic of tabetic arthropathies. Enchondral ossification of the joint cartilage can, however, be resumed only in those places where proliferative changes in the cartilage have already taken place. Cartilaginous proliferation is, therefore, a *conditio sine qua non*. The proliferation of the cartilaginous cells is followed by the invasion of well vascularized marrow spaces into the subchondral bony lamella and the zone of preparatory calcification, and, frequently, even into the deep noncalcified layers of joint cartilage. If proliferation takes place in noncalcified cartilage which is in firm union with the deeper calcified layers, the increase of volume can lead only to thickening of the joint cartilage; the cell columns become elongated and thinned out and expand only in one direction. If, however, the solid union between noncalcified and calcified cartilage has become loosened by the invasion of bone marrow in the deeper layers, then the proliferation of the noncalcified layers of cartilage leads not only to an increase in thickness but also to an increase in total area, and the resulting disproportion in size between the proliferative cartilage and the subchondral zone finds expression in an implication or coarse waviness of the joint surface.

3. Moritz, A. R.: Virchows Arch. f. path. Anat. 267:746, 1928.

In one case (tabetic arthropathy with fracture of the neck of the femur),<sup>2d</sup> the active variety of fold formation led to such marked unevenness of the joint surface that several pleats could be seen easily with the naked eye. Figure 9 is a photomicrograph (low power) of the head of the femur, with the characteristic changes. There is elongation of the cell columns and mucoid degeneration of the cartilaginous ground substance—the two most important changes in tabetic joint cartilage. Half of the cartilaginous folds are directed toward the joint cavity, and the alternate half toward the subchondral spongy bone. One might think that one was dealing here with passive pleating of the joint cartilage which had been displaced by trauma, as in the group previously mentioned. But this supposition may be excluded because: (1) the proliferating and thickened cartilage blends gradually with the old cartilage,



Fig. 9.—Active process of fold formation in a case of tabetic arthropathy. The subchondral bony lamella with parts of the zone of preparatory calcification is preserved; the noncalcified cartilage is invaded by marrow spaces and shows active proliferation of its cell groups with consequent fold formation. Cartilage formation is present in the pannus on the joint surface.

which is firmly united with the subchondral spongy bone, the cells of which are perfectly quiescent, and (2) the subchondral bony lamella remained in place, together with parts of the zone of preparatory calcification, thus forming a good landmark to show where the joint cartilage once ran and how it became elevated and formed folds to accommodate its lateral expansion (*Flächenwachstum*).

There is another feature of this case which shows clearly that the unevenness of the joint surface is due to proliferation of the cartilage. In one place the deeper layers have been resorbed by bone in such a way that the cartilage became undermined for a considerable distance by large marrow spaces. These marrow spaces are in the stage of obliteration by the implication of the joint cartilage toward the preserved

subchondral hard substances. The marrow of the intracartilaginous marrow spaces becomes in this way compressed and transformed into dense fibrous tissue, which in some places undergoes transformation into fibrous cartilage. In no place can signs of traumatization be seen. All the changes may be explained by the high activity and the power of proliferation of the tabetic joint cartilage, with subsequent invasion by marrow spaces.

As to the practical value of these observations in the group of cases showing passive fold formation as well as in that showing active fold formation, it is clear that the process leading to such marked deformities must be detrimental to action of the joint. This is especially true in younger persons, when the underlying disease is not fatal *per se*. In cases of Perthes' disease, I found, as a rule, an early onset of hypertrophic arthritic changes in the hip joint.

#### SUMMARY

Under pathologic conditions the joint cartilage may show more or less marked plication. The process leading to this unevenness may be active or passive, the latter being by far the more frequent. Both forms result from a disproportion in the dimensions of the bony epiphysis and of its cartilaginous cover. If the total volume of the bony epiphysis becomes diminished by destructive or resorbing processes, the joint cartilage may follow the diminution of epiphyseal bone, with the formation of folds, the apexes of which are directed toward the center of the epiphysis; this is the passive type. In the active form the disproportion between cartilage and epiphyseal bone arises from augmentation of the cartilaginous volume by extensive proliferative processes in the cartilaginous tissue. By these changes the cartilage becomes too large for the bony epiphysis and forms folds, the apexes of which are directed mainly toward the joint cavity.

# HISTOLOGIC CHANGES IN THE KNEE JOINT IN VARIOUS INFECTIONS

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In previous articles we<sup>1</sup> described the changes observed in the knee joint with advancing age. It was pointed out that an increased number of anatomic alterations were seen as age advanced and that the gross and microscopic examination of these lesions showed changes characteristic of the various stages of so-called degenerative arthritis. As these observations defined the type of lesion resulting from a degenerative process, we studied the knee joints of eight patients with infections of the joint cavity in order to compare the changes in an inflammatory lesion of the joints with those resulting from a degenerative process. There were two cases of gonococcic, two of hemolytic streptococcic, two of meningococcic and one of pneumococcic, infection and one showed an unidentified gram-negative coccus causing subacute bacterial endocarditis and arthritis.

## GONOCOCCIC ARTHRITIS

CASE 1.—A man, aged 42, was admitted to the hospital with pain in the knees of eight weeks' duration. He had a similar attack of polyarthritis following gonorrhea three years before admission. On examination, he showed psoriasis, chronic gonococcic urethritis and an acute arthritis of both knee joints. Gonococci were obtained from the urethral discharge, and the complement-fixation reaction of the blood was positive with a gonococcus antigen. Five days after admission he had a chill, and signs of lobar pneumonia developed in the lower lobe of the right lung. During the course of the pneumonia the psoriasis disappeared. He failed progressively and died six days after the onset of the pneumonia.

Necropsy showed the lesions of a typical pneumococcic lobar pneumonia and the following changes in the joints: On opening the knee joints there was no excess fluid. The patellas appeared normal. The femoral and tibial condyles were

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1. Keefer, Chester S.; Parker, Frederic, Jr.; Myers, Walter K., and Irwin, Ralph: Arch. Int. Med. **53**:325, 1934; Parker, Frederic Jr.; Keefer, Chester S.; Myers, Walter K., and Irwin, Ralph: Arch. Path. **17**:516, 1934.

likewise smooth and showed no areas of destruction, although the thickness of the cartilage over the lateral condyles of the tibia was decreased. The synovia appeared somewhat thickened; the surface was smooth except in several small areas where it was dull and irregular.

Microscopic examination of the synovia from the right knee joint showed a slight irregularity of the surface where the superficial layer of cells varied in thickness. There were no definite papillary projections. In the connective tissue layer there was a marked infiltration with polymorphonuclear leukocytes, lymphocytes and macrophages with an intense congestion of the blood vessels. An occasional macrophage filled with blood pigment was seen. In several foci there was a partial loss of the superficial synovial cells with a deposit of fibrin. In places the collagen and polymorphonuclear leukocytes appeared necrotic. A careful search of the tissue, stained for bacteria, failed to reveal their presence.

The synovia from the left knee joint showed an infiltration with some polymorphonuclear leukocytes, lymphocytes and macrophages. In some areas the connective tissue was edematous; in others it was dense. Over the surface the synovial cells were thin and flat. The vessels were prominent and seemed to extend closer to the surface than usual. No organisms were demonstrated.

In this case, the principal lesion was in the synovial tissues and was characterized by an inflammatory reaction which had extended to the surface of the synovia in some places. In other words, it was a synovitis beginning in the synovial connective tissues.

The following case was a much more advanced stage of the same type of infection:

CASE 2.—A man, aged 50, was admitted to the hospital on account of pain and swelling in the left knee joint. He had had a chronic urethritis for a number of years. On examination, he showed a bilateral catarrhal conjunctivitis, slight fever and an arthritis of the left knee joint. This joint was swollen, painful and tender. The overlying skin was red and the surface temperature increased. The patella was elevated, and there was definite fluctuation of the joint capsule. The urethral discharge showed many gonococci, and fluid aspirated from the left knee joint had the characteristics of an exudate; gonococci were seen on smear and grown in culture. During the period of observation the patient continued to have high irregular fever and leukocytosis; and, in spite of repeated aspirations, the fluid continued to accumulate in the joint. The blood cultures were sterile. Within two weeks, 1,065 cc. of purulent fluid was removed from the left knee joint. The patient became progressively weaker and died one month after admission.

Necropsy showed a bilateral hydrothorax and a purulent arthritis of the left knee joint. Examination of this joint showed that it contained an excess of muco-purulent exudate. The bursae communicating with the joint were likewise filled with purulent material from which gonococci were recovered. The proximal tibio-fibular joint was likewise filled with a similar exudate. The synovial membrane was swollen, thickened and markedly injected. The surface was rough, dull and covered with exudate. The articular cartilage of the patella showed fibrillation of the cartilage of the median horizontal facets such as is seen in many patients of this age. On the articular surface of the femur there were erosions on the patellar groove and on the lateral and medial condyles. The tibia showed thinning of the cartilage over the areas of pressure. In no place was there evidence of destruction of the cartilage by the inflammatory process in spite of the fact that the infection had been present for several weeks.

Histologic examination of the synovial membrane revealed that the superficial synovial cells were absent and that they had been replaced by granulation tissue containing numerous lymphocytes, polymorphonuclear cells, macrophages and plasma cells. In the deeper layers of the synovia there was a perivascular infiltration of lymphocytes. The sections stained for bacteria showed numerous gram-negative cocci which were undoubtedly gonococci, since this organism was recovered from the tissue on culture.

The changes observed in this knee joint were those of an acute synovitis without any striking alteration in the cartilage or bone.



Fig. 1 (case 1).—The synovial membrane from a patient with gonococcal arthritis showing the inflammatory reaction beneath the superficial cells of the membrane ( $\times 170$ ).

In these two cases the reaction differed in intensity. In the first case, no gonococci were recovered from the joints at necropsy, and no organisms could be found in the stained sections. In the second case gonococci were grown from the exudate and found with ease in the stained sections of the synovia. These two cases represent what is often seen clinically, namely, a type of arthritis associated with definite gonococcal infection of the urethra or other parts of the genito-urinary

tract in which gonococci either are not found or are rarely grown from the joint fluid, and a second type in which there are many gonococci in the fluid. As a rule, as far as complete recovery from the joint disorder is concerned, the outlook is better in the former and the reason seems plain from the character of the histologic changes in the joints. That is to say, the inflammatory lesions are less intense and are characterized by an infiltration of polymorphonuclear leukocytes, lymphocytes and plasma cells. The surface



Fig. 2 (case 2).—Synovial membrane showing infiltration with leukocytes and complete destruction of the superficial synovial cells ( $\times 95$ ).

layer of the synovia remains intact and shows no areas of destruction (fig. 1). This picture contrasts strikingly with the observations in case 2 in which the synovial lining was completely destroyed and replaced by granulation tissue containing lymphocytes, polymorphonuclear cells, macrophages and plasma cells (fig. 2). Accompanying these changes were areas of perivascular infiltration of lymphocytes. In other words, in the first case the prominent lesions were beneath the surface of the synovia, whereas in the latter they extended to the surface and produced complete destruction of the superficial cells.

## STREPTOCOCCIC ARTHRITIS

CASE 3.—A white woman, aged 76, was well until three weeks before admission when she fell and injured her right knee. This was soon followed by pain and swelling which continued until admission. On examination it was found that she had fever, moderate senile kyphosis and generalized arteriosclerosis. The right knee was swollen, hot, tender and painful. Otherwise, no striking abnormalities



Fig. 3 (case 3).—Gross specimen of knee joint from a patient with streptococcic osteomyelitis and arthritis.

were found. The temperature varied between 98.6 and 102 F.; the patient gradually became stuporous and incontinent; signs of bilateral bronchopneumonia developed and death occurred five days after admission.

The white cell count varied from 6,700 to 16,100 per cubic millimeter with 82 per cent polymorphonuclear leukocytes; the blood culture was positive for *Streptococcus haemolyticus*.

Necropsy showed a septic arthritis of the right knee joint, osteomyelitis of the right femur, bilateral bronchopneumonia and generalized arteriosclerosis. Cultures of the blood and right knee joint showed hemolytic streptococci.

On opening the right knee joint the synovia appeared dull gray and projected over the edge of the patella. The fluid was mucopurulent and contained many micro-organisms. The patella showed marked erosion of the cartilage so that it was thinned over the lateral facet. The same was true of the cartilage over the median facet. The articulating surface of the femur showed thinning of the cartilage, and on the lateral surface of the patellar groove there was an irregular area measuring 1.5 by 1.5 cm. which extended through the subchondral bone into the marrow. There was a focus of osteomyelitis that had perforated into the joint cavity. The synovial membrane about the femur was greatly thickened and was continuous with granulation tissue which extended over the surface of the bone.

The articular surfaces of the tibia showed marked thinning of the cartilage with areas of irregularity, especially over the points of greatest pressure. These gross changes are shown in figure 3.

The left knee joint showed no signs of infection, but there were erosions over the middle portion of the patella, the patellar groove of the femur and the articulating surfaces of the tibia.

Microscopic examination of the synovia showed that in places there was a complete destruction of the membrane down to the fat tissue and the large nerves. The exudate was composed mostly of polymorphonuclears, with some lymphocytes, macrophages and plasma cells deep down in the tissue. The endothelium of the blood vessels showed proliferation.

The bone showed many abscesses in the marrow. While the bone was not apparently necrotic, it was being dissolved. This process involved the cartilage also. At one point the cartilage had almost completely disappeared, and the process extended across the surface with diminishing intensity; that is, there were necrosis and disappearance of cartilage and infiltration with many polymorphonuclear leukocytes; then solution of the superficial cartilage with invasion of the necrotic cells by polymorphonuclear cells; then masses of bacteria and no leukocytes, with necrosis of adjacent cartilage cells. The organisms were shown by culture to be hemolytic streptococci. They were most numerous in the advancing portion of the lesion (fig. 4).

This patient had an osteomyelitis of the lower end of the femur which had perforated into the right knee joint and produced an infection of the synovia with destruction of cartilage and bone.

CASE 4.—In a young man with a deformity resulting from a healed poliomyelitis a trophic ulcer developed about the left knee joint. This became infected with hemolytic streptococci, and the infection spread into the joint cavity by direct extension. It became necessary to amputate the leg above the knee.

The knee joint showed that the condyles of the femur were small and not well developed. The depth of the patellar groove was shallow. The synovial membrane was thickened and covered the posterior aspect of the patella almost completely. It was also adherent to the lateral condyle of the femur and extended into the joint cavity at this point. Over the articular surface of the median condyle of the femur there was a superficial erosion of the cartilage with irregularity of the surface due to fibrillation. Over the corresponding articular surface of the median tibial condyle there was erosion of the cartilage.

The semilunar cartilages were small and irregular, showing numerous irregularities and tears with fibrosis and evidence of a process of repair progressing in the torn areas.

The synovial membrane was thickened, and a large part of the surface of the membrane was destroyed. In areas where the synovial cells were present, the lining consisted of rather large cells which were covered here and there with fibrin. Beneath the surface there was a marked infiltration of lymphocytes and macrophages containing hemosiderin. The connective tissue was vascular, appeared young and was infiltrated with a considerable number of leukocytes. In some

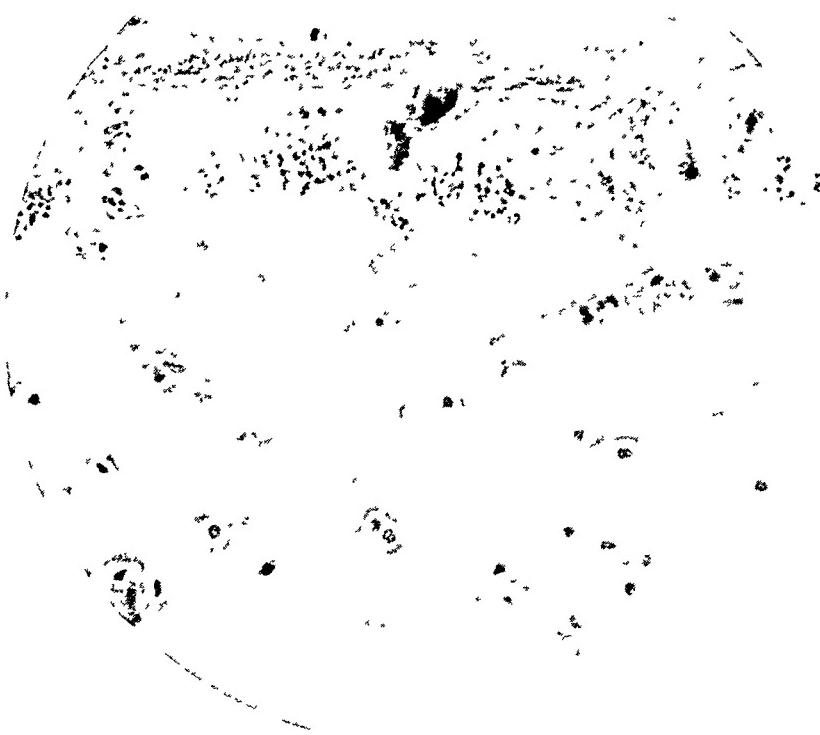


Fig. 4 (case 3).—The superficial layer of cartilage shows beginning destruction due to the advancing streptococcic infection ( $\times 95$ ).

areas the synovial lining was completely destroyed and replaced by a dense layer of granulation tissues infiltrated with numerous plasma cells and polymorphonuclear leukocytes. Beneath these areas there was a marked perivascular infiltration of lymphocytes and plasma cells. The connective tissue also appeared young and was edematous. The capsule did not appear abnormal.

#### MENINGOCOCCIC ARTHRITIS

There were two patients with meningococcic meningitis and arthritis; one was 34 years of age, and the other, 42. The clinical history and anatomic findings in the knee joints were as follows:

CASE 5.—A white man, aged 34, was admitted to the hospital with headache and signs of meningitis. The spinal fluid showed the characteristics of purulent meningitis, and meningococci were seen on smear and grown by culture. Thirteen days after admission the patient complained of pain, tenderness and slight swelling of the left elbow joint. There was no redness. At no time did he complain of pain in the knee joints. He died twenty-one days after admission. The elbow was not obtained at autopsy, but both knee joints were opened and material removed for examination.

The right knee joint was normal in appearance and contained no excess fluid. The surfaces of the joints appeared smooth and glistening. The median surfaces of the patellas were smooth; the lateral surfaces showed an elevation of the border which was due to a thinning of the articular surface of the cartilage, pressing the cartilage at the edge outward. The synovial membrane extended over the edge as papillary projections. The femoral surface showed that the lateral condyle was normal; the medial condyle showed a defect 2 by 3 mm. and corresponded to the area which came in contact with the intercondyloid tubercle of the tibia. The tibial surface was smooth; the capsule was not thickened. In no area was there evidence of destruction of bone or cartilage by inflammation.

Microscopic examination of the synovia showed that the surface was intact and smooth. The tissue between the superficial layer of synovial cells and the deeper layers of the synovia showed a marked infiltration with polymorphonuclear leukocytes, a few lymphocytes and occasional mast cells. There was one thrombosed blood vessel with an infiltration of polymorphonuclear cells, and in another area a perivascular infiltration with polymorphonuclear leukocytes without fibrin or necrosis. A section stained for bacteria by the MacCallum-Goodpasture method showed beneath the superficial layer of cells focal collections of polymorphonuclear leukocytes, macrophages and numerous gram-negative diplococci, mostly extracellular.

In other words, micro-organisms could be demonstrated in the connective tissue of the synovia, and there was the reaction of acute inflammation about the bacteria. In this case, the infection was so slight that it did not extend to the surface cells and cause an effusion into the joints.

CASE 6.—A white man, aged 42, was admitted to the hospital with a sore throat, pain in the neck and in both knees, numbness of both legs, general malaise and prostration. These symptoms had been present for two days and were ushered in abruptly with a chill. On admission the patient was conscious but extremely ill. The temperature was 104.4 F.; the white cell count, 26,500 per cubic millimeter. Signs of meningitis were present, and the cerebrospinal fluid was characteristic of a purulent meningitis. Meningococci were grown from the blood and spinal fluid. The patient died on the third day of his illness.

The right knee joint did not contain an excess of fluid, and the synovia appeared smooth without evidence of acute inflammation. At the border of the patella there was a tendency for the synovia to extend over the edge and encroach on the articular surface. The femoral and tibial condyles were smooth and, aside from a slight thinning of the cartilage over the articular surface of the tibia, no abnormalities were observed. The left knee was similar in all respects to the right. On microscopic examination the synovial membrane showed two types of lesions. The first was characterized by changes confined entirely to the connective tissue beneath the thin layer of synovial cells. The surface of the synovial membrane was smooth and intact, the cells appearing normal. In the connective tissue there were numerous areas showing infiltration with polymorphonuclear cells, a

few lymphocytes and mast cells. These cells were concentrated about the smaller blood vessels just beneath the surface and between the strands of connective tissue. A few of the areas of infiltration were situated quite deep in the tissue, and here again the cellular infiltration was noted, especially about the blood vessels.

A more advanced stage of this process was noted in some of the sections. The superficial synovial cells appeared swollen and pale and, in many places, necrotic. In other areas, they had disappeared entirely, leaving a surface composed of granulation tissue and a thick layer of cellular exudate composed of polymorphonuclear

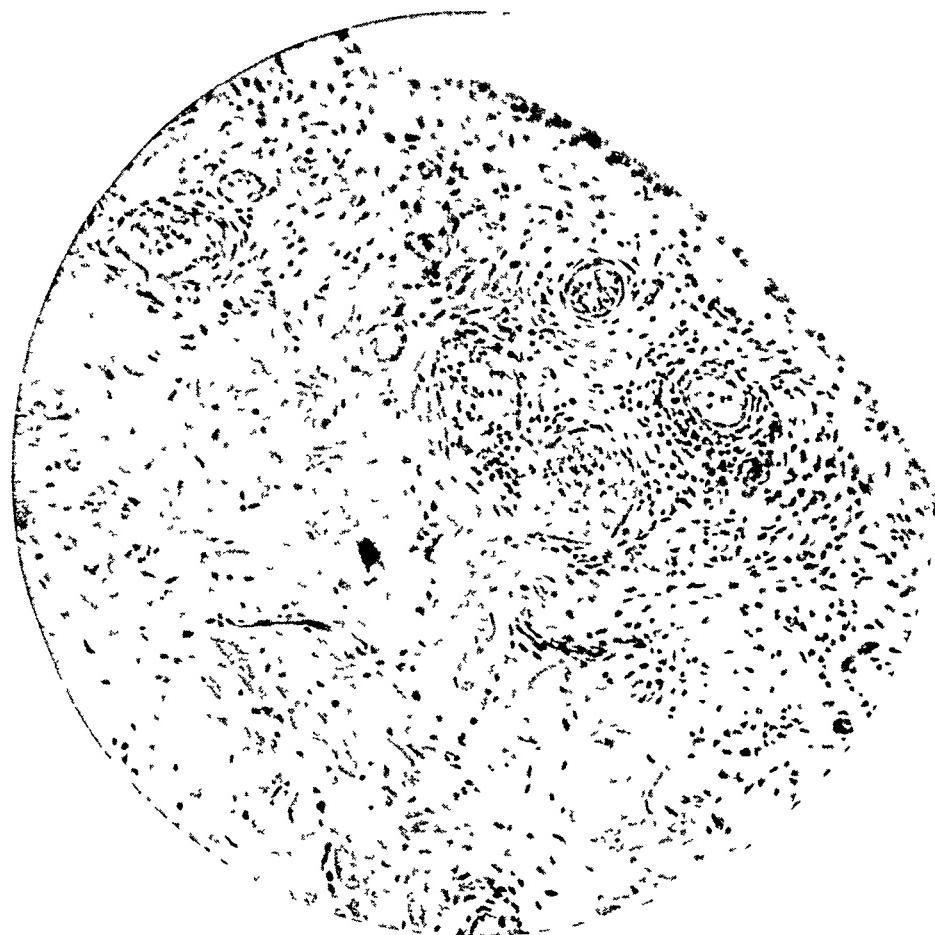


Fig 5 (case 5).—Synovial membrane from a patient with meningococcic arthritis showing the early lesions with focal collections of cells about the blood vessels and in the connective tissue ( $\times 170$ ).

leukocytes. At this stage the perivascular arrangement of the leukocytes was not striking, although here and there this could be seen. In spite of the intensity of the reaction at the surface, it was almost entirely limited to this location and did not extend far into the capsule. There were collections of cells here and there quite deep in the synovia, but they were few. A stain showed a number of diplococci in the area showing the inflammatory reaction.

The course of events in meningococcic infection of the synovia is as follows: The organisms reach the synovia by way of the blood stream

and are deposited in the connective tissue beneath the surface of the synovial membrane. This is followed by collection of polymorphonuclear cells, lymphocytes and plasma cells, especially about the blood vessels and between the strands of connective tissue. At this stage the synovial cells on the surface remain intact. Later as the infection progresses there is a disintegration of the synovial cells, and the surface is replaced by a layer of granulation tissue and numerous polymorpho-



Fig. 6 (case 5).—Synovial membrane from a patient with meningococcic arthritis showing infiltration of superficial areas with polymorphonuclear leukocytes, lymphocytes and plasma cells ( $\times 170$ ).

nuclear leukocytes. The process does not extend deeply into the capsular tissue. The essential lesion is then one of an acute synovitis (figs. 5, 6 and 7).

It has been noted previously that pains in the joints are extremely common in both the acute and the chronic form of meningococcic sepsis, with or without meningitis. The clinical features of this type of infection of the joint have been described by numerous writers. As

a rule, arthritis occurs in about from 4 to 7 per cent of the cases of meningococcic meningitis. Herrick and Parkhurst<sup>2</sup> divided the cases into three groups. In type A they included the cases in which polyarthritis was a feature of meningococcic sepsis at the onset of the disease and usually disappeared after the third day of the illness. Associated with the pains in the joints was the characteristic rash of meningococcic sepsis. The pains in the joints were usually accompanied by

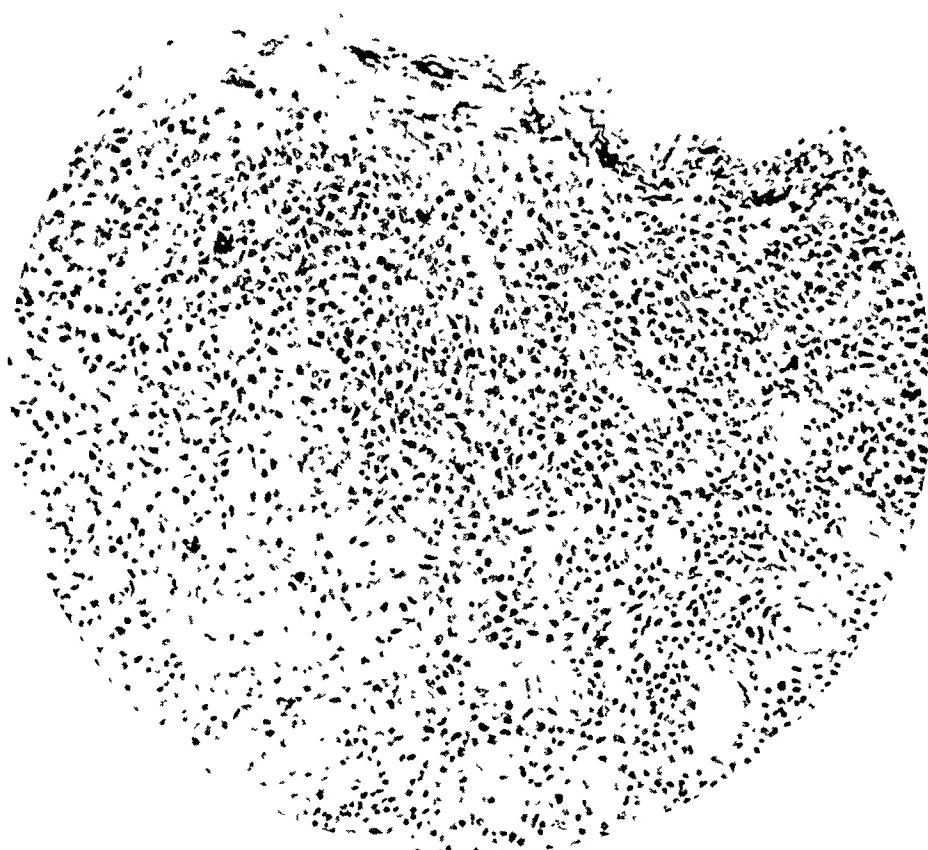


Fig. 7 (case 6).—Synovial membrane showing extensive inflammatory reaction with infiltration and destruction of the superficial areas ( $\times 170$ ).

tenderness but no swelling. It was not possible to obtain fluid from the joint for examination owing to an absence of an increased amount of fluid. In some of their cases meningitis did not appear, but other metastatic lesions such as panophthalmitis, epididymitis or pericarditis were observed. Sometimes the blood culture was positive, and if so the mortality was high. As many patients with pains in the joints

2. Herrick, W. W., and Parkhurst, G. M.: Am. J. M. Sc. **158**:473, 1919.

have a hemorrhagic rash, Herrick and Parkhurst postulated that the pains are probably due to hemorrhage into the joints and joint tissues. In case 2 of this study the pains in the joints were undoubtedly similar to those just described; the blood culture was positive, and the patient died on the third day. There were no hemorrhages into the joints or joint tissues, but the pain was without doubt due to the inflammatory lesions in the synovial tissues where the organisms were located.

In the second group, called "B" by Herrick and Parkhurst, the joints, usually the large ones, were the site of a suppurative process which appeared about the fifth day of the illness. This was characterized by pain, swelling and effusion into the joints which had the characteristics of an exudate and contained meningococci in about one third of the cases. The lesion was thus metastatic, such as occurred in the first group, with the difference that it was usually a more severe infection of the joint and produced effusion. Recovery was usually complete in about from one to four weeks, although ankylosis occurred occasionally when there was necrosis of the bone.

The third type of pain in the joints described by Herrick and Parkhurst was the arthralgia of serum sickness which appeared in some of the patients following the administration of therapeutic serum.

It is plain from the previous clinical descriptions and from our histologic observations that meningococcic arthritis is a metastatic lesion involving first the deeper synovial tissues. Later the infection invades the superficial cells with effusion of fluid into the joint cavity and varying degrees of destruction of the cartilage. It is, indeed, essentially a metastatic acute synovitis.

#### ARTHRITIS OF SUBACUTE BACTERIAL ENDOCARDITIS

Pains in the joints occurring during the course of infective endocarditis of the subacute variety are far from being uncommon. Curiously enough, we have been unable to find records of the anatomic changes that may be seen. This is probably due in part to the fact that the joints do not suppurate and are, therefore, not examined post mortem.

In the following case of subacute bacterial endocarditis due to an unidentified gram-negative coccus, there were pains in the joints during life, and slight changes in the synovia were observed at necropsy.

CASE 7.—A man, aged 25, complained of dyspnea on exertion. Five months before admission he had pains in the knee joints suggestive of mild rheumatic fever. Otherwise his past history was irrelevant.

On examination it was found that he had fever, leukocytosis, the local and peripheral signs of aortic insufficiency, splenomegaly, clubbed fingers and anemia: a blood culture was positive for an unidentified gram-negative organism, and slight signs of congestive cardiac failure, albuminuria and hematuria were present. There

was slight tenderness over the joints on palpation; otherwise the findings were unimportant. The anatomic diagnosis was: vegetative endocarditis of the aortic valves, jaundice, chronic passive congestion of the viscera, infarcts of the spleen and kidney, acute nephritis, focal suppurative meningitis and subacute synovitis.

The knee joints were opened and showed no excess fluid.

A microscopic examination of the synovia showed many papillary projections, some large with apparent reduplication of the synovial lining cells and some perivascular infiltration of lymphocytes; there were also masses of fibrin at points

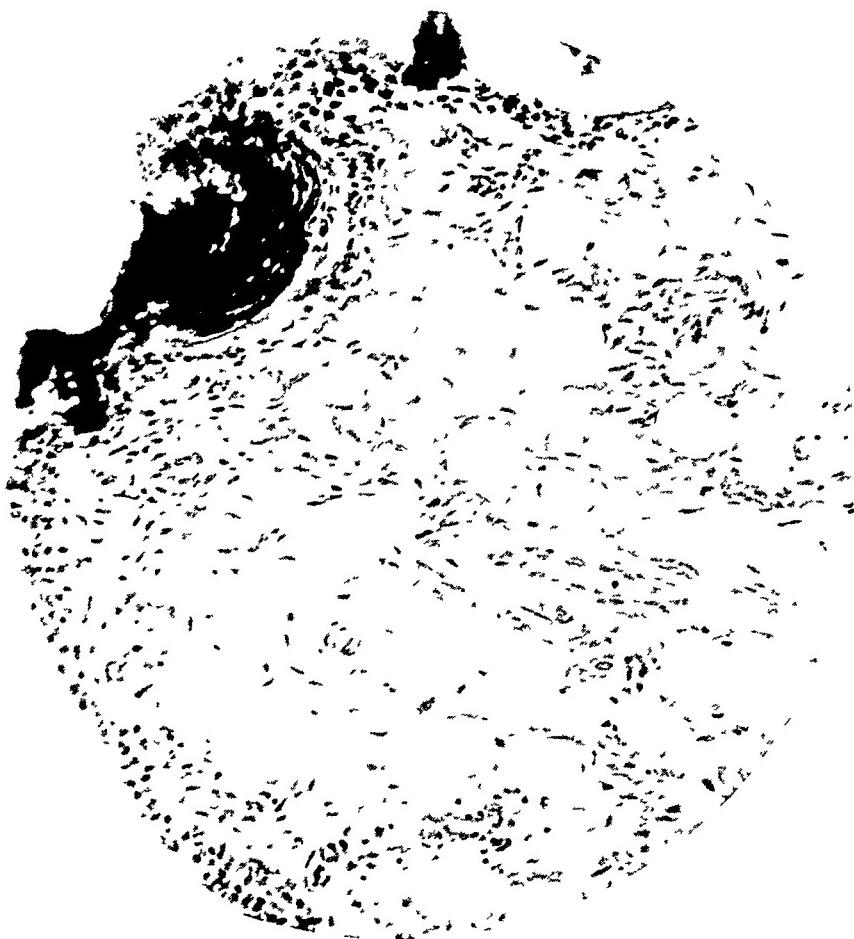


Fig. 8 (case 7).—Synovial membrane from a patient with subacute bacterial endocarditis, showing thickening of the synovia with small masses of fibrin on the surface. There is little reaction in the connective tissues ( $\times 170$ ).

on the surfaces of the projections. The fibrin looked fairly old in some areas, and leukocytes occurred in and around it. In a few projections some macrophages with blood pigment were present. In the synovial connective tissue there were scattered lymphocytes and a few mast cells. Other sections showed the same changes but to a much less marked degree.

The essential lesion was in the synovial connective tissue with perivascular infiltration of lymphocytes. The surface of the synovia was irregular in some areas and showed some masses of fibrin. On the whole, however, the surface cells were intact. The changes are shown in figures 8 and 9.

## PNEUMOCOCCIC ARTHRITIS

CASE 8.—F. P., a white man, aged 52, was admitted to the hospital with pneumococcus type I lobar pneumonia of nine days' duration. The left knee joint had been fused ten years before on account of an arthritis of unknown cause. He was cyanotic and jaundiced. There were signs of solidification of the right lung and of peritonitis. The left elbow joint was swollen, painful and tender; when a needle was inserted into the cavity, pus containing pneumococcus type I was

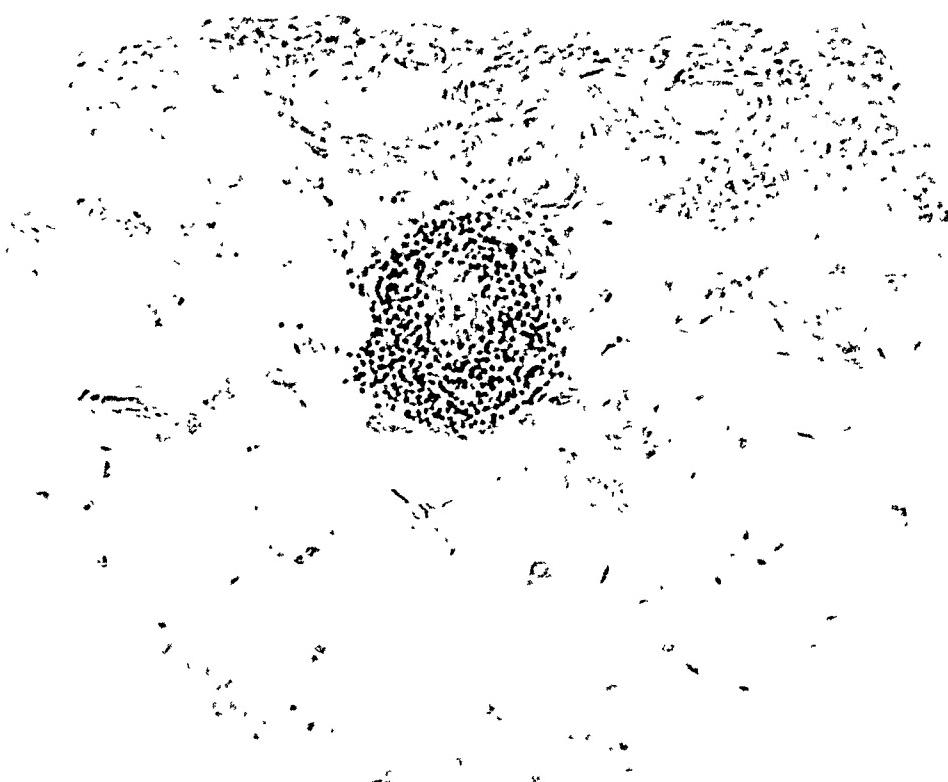


Fig. 9 (case 7).—Synovial membrane showing perivascular infiltration of lymphocytes. The patient had progressive subacute bacterial endocarditis with pains in the joints ( $\times 170$ ).

removed. The white blood cells numbered 16,200 per cubic millimeter. Death occurred on the tenth day of the disease. The clinical diagnosis was pneumococcus type I lobar pneumonia, pneumococcic arthritis of the left elbow joint and peritonitis. The autopsy confirmed this diagnosis.

On opening the left elbow joint it was found that the capsule was distended with pus. The entire articular surface of the humerus was completely denuded of cartilage and appeared irregular and granular. The synovial membrane was injected, thickened and covered with a fibrinopurulent exudate. At the edges of the humerus it was adherent. The articular surface of the ulna was similar in

appearance to that of the humerus; that is, the cartilage was completely destroyed leaving the underlying bone exposed and covered with exudate. The head of the radius was irregular, and the cartilage was destroyed.

Microscopic examination of the synovial membrane from the elbow joint showed that the synovial lining was destroyed and the surface covered with débris, leukocytes and, in places, masses of gram-positive diplococci. In the synovial connective tissue there were numerous lymphocytes, some plasma cells and a number of foci containing many macrophages engorged with blood pigment. Scattered through the tissues in places were numerous gram-positive diplococci. Smaller blood vessels throughout showed swelling of the endothelium, often with a perivascular infiltration of lymphocytes and macrophages, sometimes with thrombosis. Three large vessels showed acute lesions consisting of necrosis of their walls with fibrin deposits and infiltration with numerous polymorphonuclear leukocytes and some macrophages.

For the most part, the articular cartilage was completely destroyed. In places it persisted as a thin layer, definitely necrotic. There was an occasional island of living cartilage, especially in the deeper layers.

At the articular surface, where the cartilage had been lost, the normal subchondral bony layer had also disappeared for the most part. In this region there were trabeculae of bone surrounded by pus. The trabeculae were narrowed and irregular, and were evidently undergoing solution. Nuclei were present in the greater part of this affected bone. Where the bone marrow still persisted, osteoclasts were attacking the bone. In the deeper portions of the epiphysis, the bone as a whole was unaffected in spite of the numerous abscesses in the marrow. In one area where the cartilage and subchondral bone persisted there was a depression in the line of the joint with thickening and distortion of the bony trabeculae.

Scattered throughout the marrow were numerous small abscesses in which numerous cocci could be seen. Apart from these abscesses there were some areas of connective tissue proliferation, sometimes infiltrated with lymphocytes. This was especially prominent in one area near the articular surface where there was a marked depression in the surface of the joint.

Both knee joints were examined. The left showed a complete bony fusion, the result of an operation done ten years before. Neither gross nor microscopic examination of the tissues of the right knee showed any abnormalities.

The sequence of events and the pathologic changes in the joints in pneumococcic arthritis when the lesion begins as a blood-borne metastasis is as follows: At first there is an intense inflammatory reaction of the synovial membrane, starting beneath the surface and extending into the cavity of the joint. This is soon followed by destruction of the overlying cartilage and, finally, of the bone.

In most of the reported cases of pneumococcic arthritis the disease has appeared during or following a pneumococcic sepsis, the most common being pneumococcic lobar pneumonia. The arthritis is more often monarticular than polyarticular; the large joints are involved more frequently than the smaller ones, and the lesion is almost invariably suppurative. Associated with the arthritis there may be other metastatic lesions such as meningitis or endocarditis. Their presence, of course, definitely changes the prognosis. It is well to remember that pneumococcic arthritis requires prompt action with drainage of the joint cavity.

## COMMENT

From these eight cases, certain facts are obvious. Infections of the joints occur in one of three ways: (1) extension of an infection directly into the joint cavity from an infection or wound of the skin (case 4); (2) extension of an infection from the bone directly into the joint cavity (case 3); (3) infection of the joint cavity by way of the blood stream (cases 1, 2, 5, 6, 7 and 8). An appreciation of the mode of infection will aid in an understanding of the final picture. By far the most common is the hematogenous route. This is true regardless of the infective organism, and in the cases described this was true of the meningococcic, gonococcic and pneumococcic infections. In these infections the inflammation began in the synovial connective tissue and about the blood vessels, later spreading to the synovial cells on the surface. If the lesion progressed, the cartilage and underlying bone were attacked and destroyed. If, on the other hand, the infection spread to the joint cavity from an osteomyelitis, then there was considerable destruction of both bone and cartilage and secondary involvement of the synovia, the principal lesion in the latter being localized near the surface and spreading downward rather than by the mode of progression described for involvement of the synovia from a hematogenous process.

Aside from the mode of infection there are other factors determining the final pathologic state; these will not be discussed at this time, but require consideration. They are: the type of infecting organism, the effect of pressure, the character of the cellular reaction and the presence of anti-ferment substances in the synovial fluid.

## SUMMARY

The pathologic lesions of eight cases of infective arthritis due to streptococcic, gonococcic, meningococcic and pneumococcic infections and to an unidentified gram-negative coccus are reported.

The character of the change varied with the mode of infection, which occurred in one of three ways: (1) by the blood stream; (2) by direct extension from an osteomyelitis; (3) by direct extension from the skin overlying the joint.

In the cases in which the infection of the joints occurred as a result of a hematogenous infection, the process began in the synovial connective tissue, with infiltration of polymorphonuclear, lymphoid and plasma cells about the blood vessels and between the strands of connective tissue. As the infection progressed, the synovial lining was destroyed and completely replaced by granulation tissue. Later the cartilage and bone were involved in the process and destroyed.

When the bone was involved primarily, the outstanding lesions were a destruction of bone and the overlying cartilage. The inflammation of the synovia showed a progression from the superficial to the deeper layers.

The changes were characteristic of an inflammatory process and could be readily distinguished from degeneration.

# THROMBO-ANGIITIS OBLITERANS

DISTRIBUTION OF THE LESION IN THE VESSELS OF THE LEG

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Since Buerger,<sup>1</sup> in 1908, first described thrombo-angiitis obliterans as a definite clinical and pathologic entity, affecting the arteries of the extremities, many contributions have been made to the literature on this subject. Subsequent to this date Buerger,<sup>2</sup> Allen and Willius,<sup>3</sup> Lewis,<sup>4</sup> Taube,<sup>5</sup> McGregor and Simson,<sup>6</sup> and Telford and Stopford,<sup>7</sup> to mention only a few of the many writers, have described the disease in almost all the arteries of the body.

In 1909 Buerger<sup>2a</sup> presented the results of his studies on this disease in relation to the veins. He showed that both arteries and veins were involved in the disease process, and published a chart recording the different vessels of the leg which were affected. This publication, however, did not present the distribution of the lesion within any one vessel, and a review of the literature has failed to reveal the desired information.

One of the questions that arose was: How is the disease process distributed along the course of any one vessel and how is it disseminated through the various segments of the vascular system? With the hope of obtaining some additional information in respect to these points special care was taken in the preparation and examination of the material available.

## MATERIAL AND METHOD

During the past year the vascular system of the lower extremities was available for examination in three cases. In case 1 the arteries and veins were dissected out immediately after operation (Gritti-Stokes) and fixed in solution of formaldehyde. Blocks were taken every centimeter along the course of the arteries and the microscopic sections stained with hematoxylin and eosin and by the phosphotungstic

1. Buerger, Leo: Am. J. M. Sc. **138**:576, 1908.
2. (a) Buerger, Leo: J. A. M. A. **52**:1319, 1909; (b) Am. J. M. Sc. **149**:210, 1915; (c) The Circulatory Disturbances of the Extremities, Philadelphia, W. B. Saunders Company, 1929, p. 368. (d) Buerger, Leo, and Kaliski, D. J.: M. Rec. **78**:665, 1910.
3. Allen, E. V., and Willius, F. A.: Ann. Int. Med. **3**:35, 1929.
4. Lewis, Dean: Arch. Surg. **15**:613, 1927.
5. Taube, Norman: J. A. M. A. **96**:1469, 1931.
6. McGregor, A. L., and Simson, F. W.: Brit. J. Surg. **16**:539, 1929.
7. Telford, E. D., and Stopford, J. S. B.: Brit. M. J. **2**:1035, 1924.

acid methods. In the other two cases the legs were sent to the laboratory and, in order to preserve the normal length of vessels during fixation, were treated by the following method: The skin and subcutaneous tissues of the leg and foot were removed, and the muscle planes were laid open so that when the whole limb was immersed in solution of formaldehyde the vessels were fixed *in situ*. The arteries were then dissected out along with their vena comites, accompanying nerves and perivascular tissues. After observation of the external features of the gross material, blocks were removed at intervals of 1 cm. along the course of the vessels and their cut surfaces noted. From these blocks sections were taken for microscopic study and stained by the aforementioned methods.

#### REPORT OF CASES

CASE 1.—M. S., aged 32, a laborer, a moderate smoker, born in Canada of Jewish parentage, was first admitted to the hospital on Aug. 19, 1930, with a complaint of pain in the left foot of six months' duration, with swelling and discoloration for four months. Pain was relieved by rest and heat. The patient also stated that there had been a red superficial swelling along the course of the veins of the left leg. The left posterior tibial and dorsalis pedis arteries were not palpable. The leg was swollen, blanched on elevation and assumed a mottled purple coloration when dependent. A small area of superficial venous thrombosis was present. The left leg was cooler than the right.

A left lumbar sympathectomy was performed by Dr. D. W. G. Murray, and immediately following the operation the temperature of the left leg rose 4° C. For about one year after discharge the patient was free from symptoms; then he began having recurrence of pain on exercise of the left limb. The increased warmth of the leg gradually decreased so that at the time of readmission, on July 14, 1932, it was again cooler than the right. There was a small gangrenous ulcer between the second and third toes and extending a short distance over the dorsum of the foot. A faint pulsation was felt in the dorsalis pedis artery, but the posterior tibial artery was not palpable. Several firm, cordlike structures were felt along the course of the superficial veins.

Following failure of the ulcer to heal the leg was amputated (Gritti-Stokes) on Aug. 10, 1932.

CASE 2.—C. K., aged 31, a brakeman and a moderate smoker, born in Canada of British parentage, was first admitted to the hospital on March 7, 1932, complaining of pain in the feet and calves. This disability was brought on by exertion and had gradually increased in severity for two years. At first the pain was present only after a full day's work, but at the time of admission it was felt shortly after rising. The pain was relieved by rest and heat. Seven months before admission the patient suffered an abrasion of the left great toe; this ulcerated and would not heal. On examination both feet were reddened and cool. The feet blanched when elevated and became cyanosed when lowered. A small area of ulceration was present on the dorsum of the left great toe with marked tenderness of the tip of this toe. The ulcer failed to heal in two months of general supportive treatment, and a left lumbar sympathectomy was performed, following which there was a good rise in the cutaneous temperature of the left limb. The ulcer healed; the patient was able to walk farther, and the foot was more comfortable.

The patient was readmitted two months later with the ulcer again present. There was also an increasing disability of the feet and legs, with progressive

diminution of exercise tolerance before the onset of pain. At this time the left great toe was amputated with slow but good healing of the stump.

Five months after discharge the patient returned to the hospital with the complaint of increasing pain in the right leg and foot so that his walking limit was reduced to 200 yards (182.8 meters). On examination the right leg was cooler and of a deeper color than the left, and the dorsalis pedis and posterior tibial arteries were not palpable. A right lumbar sympathectomy was performed by Dr. D. W. G. Murray, following which there were increased heat, comfort and exercise tolerance of the limb. The patient again returned to the hospital on June 21, 1932, with a small nonhealing ulcer at the site of the amputation. There was no pain in either calf; exercise tolerance was increased, and none of the peripheral vessels were palpable. The ulcer healed very slowly.

Following discharge the ulcer again broke down and became gangrenous. All methods of treatment proved unsuccessful. A Gritti-Stokes amputation of the left leg was performed on Feb. 22, 1933.

CASE 3.—R. H., aged 47, a gardener and a moderate smoker, born in Canada of British parentage, was admitted to the hospital on May 3, 1933. Two years previously he began to have pain and numbness in the left hand, beginning in the left index finger. The pain became progressively worse, and the hand and arm became cold and waxen. Three months after the onset of the symptoms a cervical sympathectomy was performed by Dr. D. E. Robertson. Following the operation there was relief of pain, with increased warmth of the arm and hand.

Eighteen months before admission to the hospital pain appeared in the right foot; this was soon followed by swelling. During the eight months previous to admission almost the entire dorsum of the foot became ulcerated and gangrenous. The patient was unable to walk or use his arms and hands without suffering from cramplike pains.

On examination the left radial artery was not palpable. The left foot and leg were swollen and edematous, with large sloughing gangrenous ulcers covering almost the entire dorsal surface of the foot. No vessels were palpable in the left leg.

On May 4, 1933, Dr. R. I. Harris performed a Gritti-Stokes amputation, with good healing of the stump.

#### PATHOLOGIC CHANGES

Although both the gross and the microscopic morbid anatomic changes of thrombo-angiitis are well known, it is not amiss to state briefly the salient points of these observations. Of the various methods of recording the results of these histologic studies a classification which divides the disease process into acute and chronic stages has been chosen. This has been done although it is realized that the lesion begins as an acute process and later progresses to the chronic stage and that all gradations between these extremes may be observed. It is believed, since the great majority of all the sections studied fall within either of these groups, that this classification is the most useful. The following is a brief description of the lesions as seen in the gross and microscopic examination of the available material.

*Gross Appearance.*—The three cases presented a marked similarity in the gross appearance of the vascular tissues. In many regions the periarterial fibrous connective tissue was greatly increased in amount and formed a firm, cordlike structure which also contained the immediately adjacent veins and nerves. The cross-section of these areas showed the lumen of the artery to be filled with a yellowish-gray, firm tissue which, when the vessel was gently milked, revealed many fine canaliculi through which sanguineous fluid escaped. The remaining perivascular fibrous tissue presented little or no increase in amount, and the artery, veins and nerves could be easily separated. The cut surface of most of the latter areas revealed

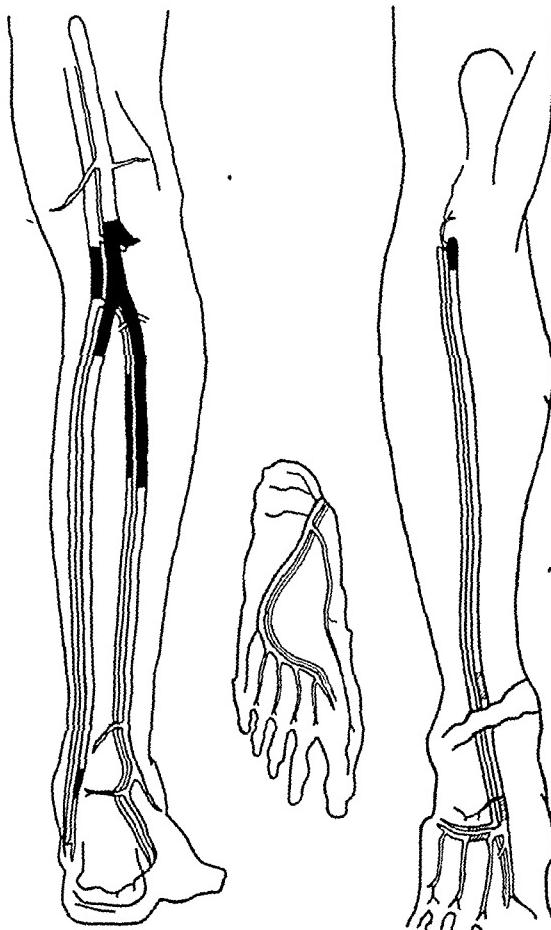


Fig. 1 (case 1).—Distribution of the chronic (solid black) and acute (barred) lesions seen in the vessels of the leg in thrombo-angiitis obliterans.

shrunken vessels with patent lumens, although in an occasional section the lumen was filled with a soft reddish-brown mass of fresh thrombus. Thus it was noted, that where the chronic arterial lesion was present the perivascular fibrous tissue was firm, tough and markedly increased in amount, while little or no change was observed about the acute lesion. Occlusion of the veins occurred with less frequency, and when present was usually associated with arterial thrombosis.

*Histopathologic Changes.*—*Acute Stage:* In this phase of the disease process there was evidence of an acute inflammatory reaction involving all the coats of the vessel. The lumen was filled with a fresh red clot, throughout which, especially in its periphery, there were small collections of leukocytes. The cellular infiltration

tion of the adventitia and perivascular tissue was chiefly of the polymorphonuclear variety, and these cells were concentrated about the small arterioles and venules; at times the reaction was granulomatous. The pathologic changes seen in the veins were similar to those observed in the arteries. The acute lesions of both artery and vein occupied but short segments of the vessels, and when present, were only 1 or, more rarely, 2 cm. in length. It was also noted that in the acute lesions of the veins, in which the histologic structures were still discernible, the formation of a thrombus was usually in relation to the valvular cusps.

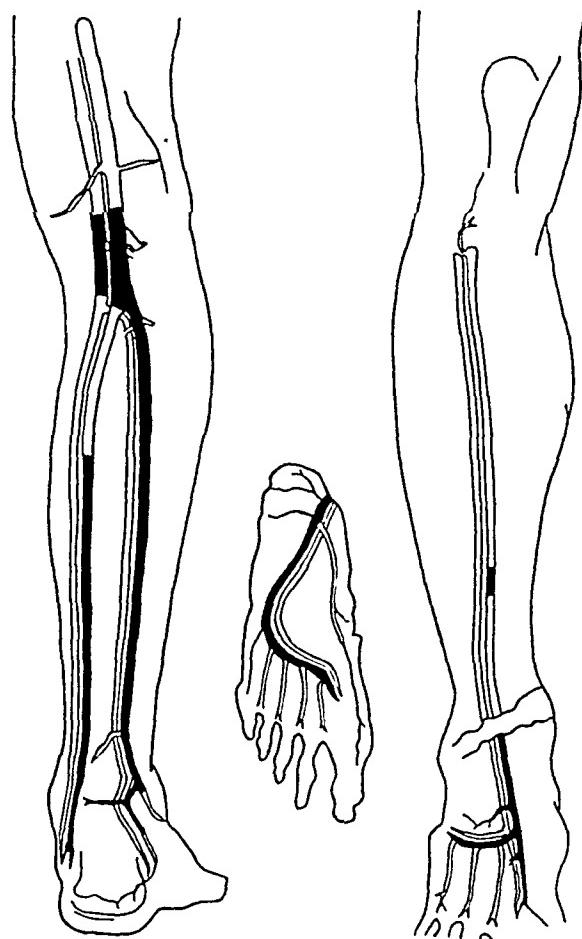


Fig. 2 (case 2).—Distribution of the chronic (solid black) lesions seen in the vessels of the leg in thrombo-angiitis obliterans. No acute lesions were present in this case.

**Chronic Stage:** Vessels showing this lesion presented the typical picture of recanalization of an organized blood clot with blood sinuses irregularly coursing through it. In this organized thrombus, especially about the sinuses, were many inflammatory cells, chiefly lymphocytes with a few polymorphonuclear leukocytes and endothelial cells. A few lymphocytes and polymorphonuclear leukocytes were scattered through the media of most of the involved areas and especially about the vasa vasorum. The perivascular tissue consisted of moderately dense fibrous tissue surrounding the artery, veins and nerves, with many leukocytes scattered throughout. The venae comites, when affected by the chronic stage of the disease, presented a picture quite similar to that described.

*Distribution in the Vessels of the Leg.*—On microscopic examination sections from each preparation showed one artery and two or more veins. Since it was obviously impossible to chart all these vessels, only the larger of the venae comites was described. In many cases, however, the condition of one or more of the smaller vessels differed markedly from that of the one recorded. For example, though the larger vein contained an old chronic obturating mass, the other vessel or vessels were patent, or filled with a fresh red thrombus, or contained a type of lesion similar to that of the larger vein. Charts 1, 2, and 3 give a diagrammatic representation of the distribution of the lesions found in the three cases.

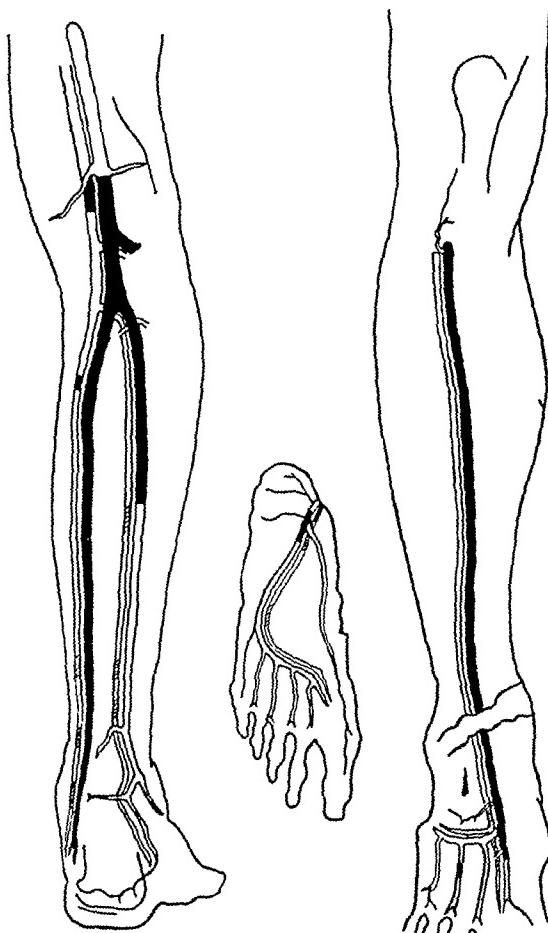


Fig. 3 (case 3).—Distribution of the chronic (solid black) and acute (barred) lesions seen in the vessels of the leg in thrombo-angiitis obliterans.

Case 1: The lumens of the lower portion of the popliteal, proximal 4 cm. of each of the anterior and posterior tibial, and upper half of the peroneal arteries were filled with masses of recanalized fibrous tissue (chart 1).

No other chronic arterial lesion was found except a small area in the posterior tibial artery at the level of the ankle joint. There were small patches of recent red thrombi in the distal part of the anterior tibial artery and in the midportions of the dorsalis pedis and arcuate arteries. Except for these regions the arteries presented little evidence of pathologic change.

The distal two thirds of the upper half of the peroneal and the lower portion of the popliteal vein were sites of the chronic stage of the disease. The only

locus of fresh thrombosis and active reaction in the veins of this leg was one small patch opposite a similar lesion in the arcuate artery.

Case 2: In this leg the popliteal, peroneal, dorsalis pedis, arcuate, distal three fourths of the posterior tibial and plantar branch of the posterior tibial arteries were all occluded by recanalized fibrous tissue typical of the chronic stage of thrombo-angiitis obliterans (chart 2). The proximal quarter of the posterior tibial and the whole extent of the anterior tibial arteries were patent, except for a small area of old thrombus formation in the upper portion of the distal third of the latter.

There were no acute lesions, and the veins were all patent except the popliteal, which was occluded by a plug of recanalized fibrous tissue having the typical appearance of the chronic stage of this disease.

Case 3: The popliteal, posterior tibial, anterior tibial, dorsalis pedis and upper half of the peroneal arteries all presented the typical appearance of the chronic lesion (chart 3). The remaining portions of the main arteries were clear, except for a small area of old thrombus formation in the proximal portion of the plantar branch of the posterior tibial artery.

The proximal 4 cm. of the popliteal vein and a small patch in the posterior tibial vein presented lesions similar to that seen in the adjacent arteries.

The venae comites of the distal halves of the posterior tibial and peroneal arteries each contained three small patchy areas of recent red thrombus and there were two similar lesions in the veins adjacent to the plantar branch of the posterior tibial and dorsalis pedis arteries.

#### DISSEMINATION OF THE LESION

The sections were examined microscopically with a view to determining, if possible, any evidence indicative of the method of spread of the disease process within the vessels of the leg. The results of this study revealed that there are at least two possible procedures: (1) by direct extension, from an initial focus, along the vessel wall; (2) by multiple acute lesions which progress to the chronic stage without spreading, the intervening spaces being later attacked by other irregularly distributed lesions.

In case 1 the most distal portion of the thrombus in the peroneal artery appeared to be the oldest lesion. At this site the occluding mass consisted of dense, mature fibrous tissue which was well canalized and contained relatively few lymphocytes. The thrombus formation in the more proximal sections consisted of progressively less mature fibrous tissue and a heavier infiltration of leukocytes. This immature type of thrombus formation filled the popliteal artery and extended into the anterior and posterior tibial vessels. From these observations it appeared as though the lesion had begun in the lower portion of the proximal half of the peroneal artery and extended upward into the popliteal artery.

In the other two cases the lesions in these vessels were of a more mature type, and the gradation from recent to old was not so apparent. Since the vessels in only three cases were examined it is difficult to draw any conclusions, although the constancy of this type of lesion, in these particular vessels, appears to be of significance. These observations, however, are strongly suggestive that the disease process begins in one region and spreads from that point.

Each of the three cases presented at least one small isolated area showing the chronic type of lesion. These observations suggest that a small patch of the acute lesion may progress to the chronic stage without spreading either proximally or distally. It is probable that as the disease progresses the intervening spaces are occupied by other irregularly distributed areas of acute thrombus formation which in their turn become chronic. This process may continue until all, or nearly all, of a vessel is occluded by the chronic lesion.

These observations suggest that either of the aforementioned two methods of dissemination of the lesion may take place within the vessels. Since case 1 presented evidence of direct spread, as described, and also patchy areas of both the acute and the chronic stage, it is probable that both types of dissemination of the disease process are present in the same case.

There is no evidence to support the theory that the primary lesion is one involving the smaller vessels of the periphery with extension from this point. Indeed, the reverse is probably true, for the gangrene of the toe in case 1 clearly antedated the acute lesions found in the vessels of the foot.

#### SUMMARY

The entire course of the vessels of the leg was examined in three cases in which a clinical and pathologic diagnosis of thrombo-angiitis obliterans was made. The three cases presented a duration of eighteen months or more, and amputation was performed only after other methods of treatment had failed. Only one of the three patients was of Jewish origin.

No one portion of an artery was unaffected in all three cases. The popliteal artery and the upper half of the peroneal artery, which were occluded by an old recanalized thrombus, were the only vessels showing a constant lesion in all three instances. A similar type of lesion involved the popliteal veins and represented the only constant localization of the lesion in the veins. The only example of (recent) thrombus formation in the arteries was found in case 1, and only one of the adjacent veins showed a similar lesion, the remainder being freely patent. In case 3 some of the venae comites contained patchy areas of acute red thrombus.

About one half of these areas were adjacent to arteries showing the chronic stage of the disease while the others were found beside arteries with little or no pathologic change.

#### CONCLUSIONS

The lesions in thrombo-angiitis obliterans may be present in any portion of any of the larger arteries or veins of the leg, though they are much more common in the arteries.

The popliteal artery, the proximal half of the peroneal artery and the popliteal vein were the only constant sites of the disease process.

The lesion may originate in any part of any artery with or without involvement of its adjacent venae comites or it may begin in any part of a vein without a similar condition being present in the immediately adjacent artery.

The lesion commences in one or more small isolated areas and later progresses to the chronic stage. The pathologic process may spread from one or more of these foci, or the intervening spaces may be occupied by irregularly scattered lesions of a similar type.

# GENERALIZED TORULOSIS ASSOCIATED WITH HODGKIN'S DISEASE

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In a recent monograph Freeman<sup>1</sup> was able to collect only forty-four instances of cerebrospinal torulosis. Generalization occurred in but three cases to which Weidman and Ratcliffe<sup>2</sup> have added an instance of extensive involvement in a chetah dying in captivity in the Philadelphia Zoological Garden. In addition to the widespread visceral involvement, the case here reported disclosed massive hyperplasia of tissue (lymph nodes?) which has never been approached in previously described cases in man, although the involvement in the chetah compared favorably with it in several respects. This report is submitted primarily, therefore, because (1) it is the fourth case with widespread visceral involvement and (2) Hodgkin's disease was associated with the torulosis (yeast cells were present in great abundance in masses of lymph nodes affected by Hodgkin's disease). It might be added that (3) the involvement of the mesenteric and mediastinal lymph nodes was so outstanding that attention must be attracted to some other portal of entry of the disease than the cerebrospinal system. The latter is commonly the only system in which the micro-organisms can be demonstrated, in which case the nasal passages are regarded as the portal of entry.

## REPORT OF A CASE

*History.*—A Negro, 18 years of age, was admitted to St. Vincent's hospital in Norfolk, Va., May 7, 1931, and died thirteen days later. He had been healthy up to three years previously, at which time the disease made its first appearance in the left axilla in the form of "hard, round lumps." Similar lesions then developed in the other axilla, the neck and both groins. They were not tender or suppurative. One year previous to admission an irritating and nonproductive cough developed, which persisted. Three months previously the face became swollen, the swelling extending down the neck on both sides. At this time dizziness and headache became so severe as finally to make the patient take to bed. In the last few days he was delirious.

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From the Laboratory of Dermatological Research, University of Pennsylvania.

1. Freeman, W.: J. f. Psychol. u. Neurol. **43**:236, 1931.

2. Weidman, F. D., and Ratcliffe, H. L.: Arch. Path., to be published.

*Physical Examination.*—This showed a greatly emaciated Negro youth with marked swelling of the face, head and neck. Questions were answered rationally, but the patient was very hard of hearing. The eyes were prominent, and both lids were swollen. The pupils were dilated and reacted sluggishly. Both disks were hazy and had engorged veins. The patient was almost totally blind, being able to see only a bright light. The ear drums appeared normal, yet hearing was reduced to that of a very loud conversational voice. There was marked congestion of the mucous membranes of the nose and throat. The neck was stiff. Lymph nodes in the neck were enlarged, varying in size up to that of a walnut. They were hard, discrete and movable, but were not tender.

The swelling of the neck did not appear to extend below the clavicles. The veins over the wall of the chest were dilated. Retrosternal dulness extended beyond normal limits on both sides. The heart appeared to be normal, and no abnormal physical signs could be elicited with respect to the lungs, except that the breath sounds were intensified. There were several enlarged lymph nodes in both axillae, one of which was as large as a hen's egg; otherwise, they presented the same physical characteristics as those in the neck.

The abdomen was scaphoid. The spleen was barely palpable. The liver was not enlarged. Many enlarged lymph nodes were present in the groins. There was no edema of the legs. Small lymph nodes could be felt in the epitrochlear and popliteal spaces.

The knee jerks were exaggerated, and there was a positive Kernig sign.

*Laboratory Examination.*—Nothing abnormal was found in the urine except a moderate amount of albumin.

The erythrocytes numbered 3,800,000, with 16,000 polymorphonuclears. The hemoglobin was 70 per cent. In the differential count there were 97 per cent polymorphonuclears, 2 per cent lymphocytes and 1 per cent eosinophils. Blood cultures were sterile on two occasions. The Wassermann and Kahn reactions of the blood were negative.

The spinal fluid was turbid and contained no globulin. Twenty-five milligrams of dextrose was found in one hundred cubic centimeters. The sediment contained many yeast cells, and occasionally a polymorphonuclear cell was noted. The Wassermann and Kahn tests gave negative results. Pure culture of a yeast organism was secured.

A node was removed from the left axilla for biopsy; scrapings from its cut surface disclosed many yeast cells. In histologic sections, however, they could not be recognized. We agree with Dr. Roache that the changes were suggestive of Hodgkin's disease, and, indeed, we should go considerably farther. The normal architecture of the node was completely obliterated by closely packed masses of lymphocytes. At scattered intervals Dorothy Reed cells were readily identifiable, and in places there were definite overgrowths of fibrous tissue. Eosinophils, however, were not found. In certain areas endothelial cells occurred, which were similar to those which will be described later in materials from necropsy and in an experiment on a cat, but which were never as conspicuous.

*Autopsy* (Mary E. Roache).—The body was that of a mulatto youth, apparently 18 years of age, highly emaciated, weighing about 65 pounds (29.5 Kg.). Rigor was absent. The intercostal spaces were sunken, the wrists prominent and the abdomen scaphoid. There was moderate enlargement of superficial cervical and axillary lymph nodes. The inguinal lymph nodes were much enlarged. Small, superficial, freely movable nodules were palpated under the skin of the arm, abdomen and lower extremities. Decubitus was present over the sacrum.

The dura was much congested and apparently thickened. The pia mater was likewise congested and exhibited areas of grayish exudate around the blood vessels, especially at the base, where the optic nerves, medulla and pons were covered in addition by thick yellowish fluid. Fluid collected from the posterior fossa contained many yeast cells, pus and other degenerated cells. The brain showed many small punctate hemorrhages through both the gray and the white matter. The medulla revealed hemorrhagic lines and minute punctate hemorrhages. Neither abscesses nor areas of softening were observed. On examination with a hand lens after fixation in formaldehyde, extremely minute cysts could be observed in the cortex of the cerebrum.

Subcutaneous fat was absent from the abdominal wall. The abdominal muscles were wasted. There was no fluid in the peritoneal cavity.

Along the mesenteric attachment of the small and large intestines were nodules from 1 to 3 cm. in diameter. In the mesentery itself there were similar masses measuring up to 9 cm. in diameter. Enlarged nodes could be felt behind the peritoneum.

The spleen was about three times the normal size. Its surface was grayish and had a thickened capsule containing two dense, firm scars. The latter, on section, were yellow, firm and fibrous. Nothing notable was observed on the surface of the section. It appeared to be infiltrated by connective tissue. The liver extended to the edge of the costal margin. It was dark red, firm and without gross lesions. The kidneys were enlarged and soft. The capsule was adherent, the surface of the kidney being torn and granular after removing the capsule. On the surface of the section the renal tissue was orange-yellow and mottled with hemorrhagic lines and dots. The pancreas was soft and flaccid. There were several nodules from 2 to 4 cm. in diameter in its head. The cut surface was pink, translucent and firm. The urinary bladder presented a few small nodules on its surface.

Covering the precordium was a wedge-shaped mass infiltrating both the pericardium and the pleura on either side. When sectioned, it measured 6 cm. in thickness; it was soft and appeared to be composed of dark red granulation tissue, with caseation and softening in the center. Direct examination of smears revealed large yeast cells and degenerated tissue cells.

The heart was moderately enlarged. The musculature did not contain nodules, nor were there any adhesions. The lungs were flaccid and partly collapsed, containing little air. Hard, enlarged lymph nodes surrounded the hilus of each lung, and similar nodules were observed under the pleura.

*Histologic Examination.*—Sections from the lungs, kidneys, spleen, lymph nodes, pancreas, cerebrum, cerebellum and spinal cord were embedded in paraffin and examined histologically.

**Lungs:** In places the walls of alveoli were irregularly fibrosed; in some regions air spaces were collapsed. At no place was there any inflammatory infiltration. Yeast cells could not be observed. The bronchi appeared normal.

**Kidneys:** The capsule appeared normal. The interstitial tissue was not increased, except in foci which will be described. The blood vessels were injected. The tubular epithelium was coarsely granular and swollen, leading to extensive occlusion of the lumens. The glomerular tufts were normal, except when affected by yeast cells, as will be noted.

**Torular Features:** These were extensive. Torula cells were observed (1) within glomerular tufts, (2) within kidney tubules and (3) in small granulomas,

which affected both the interstitial and the tubular tissue. All these changes affected the cortex almost to the exclusion of the medulla.

The glomerular tufts were frequently eight or ten times normal size, in which case they were scarcely recognizable as glomeruli. They became transformed into multilocular cysts as the result of proliferation of the fungus cells in the lumens of the capillaries, but there could not be any doubt as to such an origin

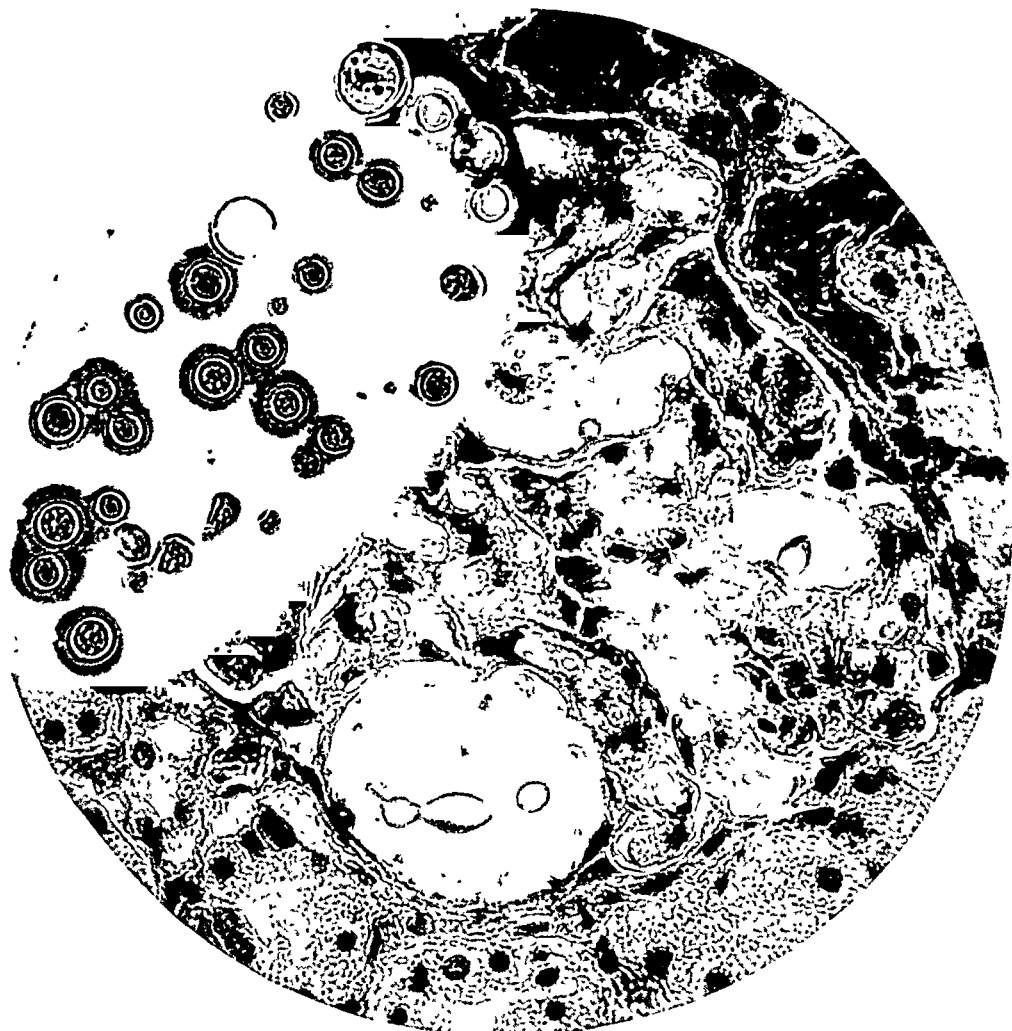


Fig. 1.—Torula cells in the capillary lumens of a glomerular tuft of the kidney; the black insert shows torula cells in culture and demonstrates the mucinoid envelop (wet india ink preparation).

of the cysts on examining smaller glomeruli in which the process was beginning. Only a small part of the contents of the cysts was comprised of the yeast cells themselves; between them were granules and minute hyaloid globules. The precise nature of the latter could not be stated with finality, i. e., whether they originated in torula cells or in degenerated tissue, but we favor the latter origin and believe that they were analogous to the "exudative" hyalin seen in glomeruli in some of the exanthems. None of the granules inclined to a bluish tint such as would indicate that the mucinoid substance observed in the interior of torular cysts at necropsy was true mucin. The yeast cells varied widely in size: smaller

ones were almost coccoid, while larger ones were larger than red blood cells. Many of the cells were budding. There was no cellular infiltration around the glomeruli.

When affecting the renal tubules, fungus cells were observed in the lumen intermixed with more or less disintegrated lining epithelial cells. In this location the lesion did not take the form of a cyst. For the most part, inflammatory reaction was absent around these foci also, but some large lymphoid cells were clustered at the margin of an affected tubule here and there. In no case, however, was such a lesion entirely invested by the infiltrate.

The third type of lesion, the granuloma, consisted mainly of small masses of yeast cells intermixed with degenerate cytoplasmic granules and nuclei of round cells, but the nuclei were not fragmented or associated with polymorphonuclears. Peripherally, the reactive cells tended to take the form of endothelioid cells, and at places the nuclei became clustered in such a way as to suggest giant cells. However, true Langhans forms were never observed, nor was there any definite lymphocytic infiltration or necrosis.

Pancreas: The interlobular fibrous tissue was slightly thickened and of old adult type. The parenchymal cells were apparently normal. The acini were poorly outlined, leading to the suggestion that they were fused (autolysis?). The islands of Langerhans were numerous, well formed and of normal size; collections of yeast cells were observed in but few. There was some budding. A solitary focus of yeast cells was also observed within one of the lobules. In neither case was there any tissue reaction around these lesions.

Spleen: There was no fibrosis of the capsule, but the trabeculae were thickened and deformed. The reticulum was also extremely thickened, particularly in the splenic nodules, where a comparatively coarse network invariably traversed them, and in places the endothelium of the sinuses had proliferated to the degree that it formed a continuous epithelium-like lining. The central arteriole could scarcely be made out, if at all. Still, the general architecture of the organ was well maintained. Lymphocytes were conspicuously scarce in the splenic pulp, but red blood cells were fairly numerous. By contrast, large monocytes suggestive of the mononuclear examples of Dorothy Reed cells were conspicuous and numerous in the sinuses, but their outlines were not sharp, and the cytoplasm was loose as though it were edematous. In addition to such hyperplastic tendencies, clusters of small (rarely larger) foreign body giant cells appeared around some minute islands of granular, almost hyaloid substance which suggested a focus either of edema or of a similar hyaloid substance. The end-result was an ill-defined granuloma unassociated with endothelioid cells, but which consisted almost entirely of the giant cells. Torula cells appeared within their cytoplasms rather frequently; more rarely they appeared independently in the centers of the granulomas. A further abnormality consisted in minute clusters of polymorphonuclears distributed numerously but at wide distances throughout the section. Eosinophils were not observed.

Yeast cells were observed in, and adjacent to, the giant cells just described and also in the lumen of smaller vessels. The inconspicuousness of central arterioles of the splenic nodules is thus perhaps explained; i. e., it is due to mycotic thrombosis at some preceding date. The coarsely granular, bronze-black pigment present in goodly quantities in the interior of the yeast cells recalled that of malaria. (Incidentally, similar grains of pigment were observed within some of the yeast cells in the pancreas.) We do not believe, however, that this was malarial pigment, as we have observed it in other cases of torulosis, including

experimental cases in rats. It is probably a product of the yeast cell itself, but may represent phagocytosed blood pigment.

The end-result of the changes just described was a definitely, uniformly and diffusely fibrosed splenic pulp, poor in lymphocytes but rich in hyperplastic endothelium and interspersed with minute granulomas comprised of small, poorly formed giant cells of the foreign body type. The picture was not quite that of Hodgkin's disease, even eliminating the granulomas. Yeast cells were not numerous, but were readily observed when searched for.

**Peribronchial Lymph Nodes:** The architecture of this organ was almost lost; its identification rested largely on the great quantity of anthracotic pigment in the fibrous parts. Only in places were there any traces of sinuses or cortical follicles. The section consisted mostly of fibrous tissue, which appeared largely as dense hyaline masses, but also as strands which infiltrated the lymphadenoid

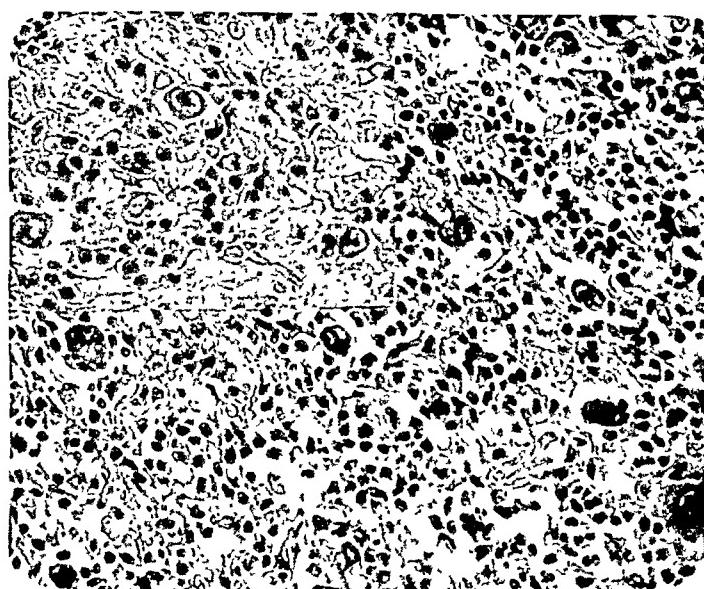


Fig. 2.—Section of the peribronchial lymph nodes; the spleen showed an identical type of change, as illustrated in the insert.

regions more or less coarsely. Elsewhere the tissue was comprised of a fibrous reticulum, which was delicate in some places and very coarse and hyaline in others. The cells within its meshes were largely lymphocytes. There were moderate numbers of endothelial cells in certain small restricted areas, but they were decidedly inconspicuous by comparison with the outspoken endothelial hyperplasia which preponderated in the mediastinal mass next to be described. Dorothy Reed cells, mostly mononuclear, but frequently classically multinucleate and with indented nuclei, were abundant. There was a rich network of blood vessels throughout, many of which were hugely dilated. Eosinophils and torula cells were not recognized.

**Mediastinal Mass, Section 1:** The changes here were of the same order as those in the peribronchial lymph node, except that the fibrosis was more extreme and the endothelial features had increased sufficiently to equal the lymphocytic ones. The Dorothy Reed cells were largely located in the more fibrous parts, much less frequently in the reticulo-endothelial ones.

Section 2: The architecture of the lymph node was scarcely recognizable; at most, only an occasional lymph sinus could be identified. Lymphocytes were scarce; when present, they were scattered loosely on the reticulum and in the sinuses. Otherwise, most of the section consisted of dense fibrous trabeculae or even expansive areas which communicated with narrower strands which represented the thickened reticulum normal in the organ. Other and extensive portions of the tissue consisted of a delicate reticulum which was not arranged according to the normal architecture of a lymph node with its sinuses, but simply as a diffusely interlacing network. On such a network great numbers of endothelial cells with broad, more or less stellate cytoplasms were observed. They were indefinitely outlined, and the cytoplasms had a rarefied appearance indicative of edema or some kindred retrogressive change. Lymphocytes were intermixed rather sparingly. In addition, other cells were abundantly present which approached decidedly the form of the Dorothy Reed cell or even attained its

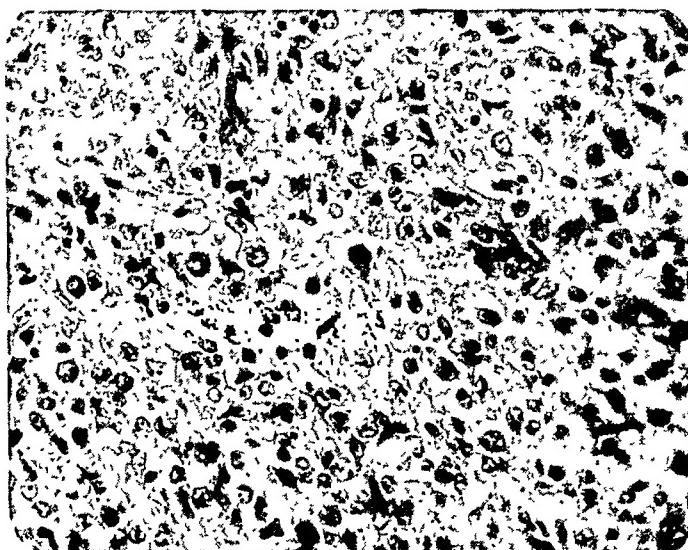


Fig. 3.—Section of the mediastinal mass, showing changes of the order of malignant lymphoblastoma.

classic form. Their cytoplasms were scanty and had frayed-out margins, and the nuclei were almost gigantic. The latter were ovoid, rich in chromatin and frequently indented. At times there were two or three such nuclei in a single cell. Capillary blood vessels were not numerous, but larger blood vessels (of venous type) were plentiful and distended. Neither eosinophils nor yeast cells were observed. Several smaller or larger areas of necrosis occurred in which the nuclei no longer took the basic stain, yet in which the general architecture just described could still be made out. Indeed, in such necrotic areas one could still identify both the older, fibrous and the younger, hyperplastic phases of the general tissue change, indicating that the necrosis was indiscriminate, involving not only the older, fibrous but also the younger, more cellular parts.

Definitely circumscribed granulomas which might be interpreted as tuberculous were not observed. However, regions of beginning necrosis were sometimes present, within which fibroblasts had become deformed in such a way as to resemble endothelioid cells, but the additional histologic criteria of tuberculosis were not present.

Section 3: In this sample of material there were none of the larger areas of hyaloid fibrous tissue, except in the thickened capsule. The latter contained, in addition, several lobules of highly degenerate, swollen skeletal muscle fibers. The nuclei of the latter were frequently of gigantic vesicular character, and their cytoplasms were highly rarefied, as though by edema. In places the edema was so extreme that in transverse section the lobule of muscular tissue might be mistaken for a lobule of fatty areolar tissue. Incidentally, there were regions deeper down in the main portions of the mediastinal mass where such cells had become enclosed and dissociated by the reticulo-endothelial cells to the extent that they became difficult to identify as muscle cells. In such position they appeared as foam cells, not so much of the xanthomatous type as of the Gaucher type. In any event, their position deep in the mediastinal mass would serve to indicate the infiltrative character of the mass, i. e., it was sufficiently extensive to enclose portions of skeletal muscle.

In this specimen also the main body of the mass consisted of a fibrous reticulum which, however, was not nearly so dense as that of the other members of the reticulo-endothelial system which have been described previously. Capillaries, however, were abundantly present on the reticulum. The cells were preponderantly endothelial (Dorothy Reed cells are included under this heading), although there were a few minor concentrations of lymphocytes in various portions of the section. Eosinophils or torula cells were not recognized, although the latter were definitely identified on direct smear made at the time of autopsy. The marked vascularity, the infiltrative character in the periphery and the large size of the nuclei of the cells in this position prompted thoughts of sarcoma. However, the adherence of most of the cells to an endothelial type and the coarseness of the reticulum prevented such a conclusion in our opinion, unless so-called Hodgkin's sarcoma be admitted to the category of the sarcoma. With such destruction of normal architecture and such a degree of hyperplasia and infiltration, the reactive processes were of an activity far surpassing any ordinary hyperplastic lymphadenitis.

Brain: Even with the loupe, minute cysts were made out in the cortex; under the microscope they were observed to be rather sharply outlined and solidly filled with yeast cells similar to those already described in the kidney. Here, too, there was no surrounding inflammatory reaction; at most, a few endothelioid cells could be discovered at the lining of the cyst. The cysts occurred far more abundantly in the sulci than over the free surface of the convolutions. Most of them were elongated, the long dimension being directed perpendicularly to the pia mater; as a blood vessel could almost invariably be distinguished in each cyst, it was obvious that the cyst had had its beginning around the blood vessels. It is notable that the nerve tissue around the cysts did not exhibit any signs of degeneration; the margins of the cysts had an almost punched-out appearance. In the meninges the fibrillar stroma had become almost entirely replaced by masses of closely packed yeast cells, within which the outlines of the arteries could still be made out.

The yeast cells occurred both free in tissue spaces and within large swollen phagocytic cells; in some cases dozens of yeast cells were packed within a single phagocyte. They were exceedingly small when compared with those seen in the kidney and spleen, and were distorted beyond recognition, probably from over-fixation. Budding or pigmentary forms could scarcely be observed. There was always a wide halo around each cell, representative of its mucinoid capsule.

Cerebellum: The meninges were thickened by enormous numbers of yeast cells, each with its halo. There were no cellular reactions against them. In only one or two of the numerous sections studied were cysts found; singularly, they were confined to the medulla.

Spinal Cord: Yeast cells were confined to the meninges. The bronze-black, spherical grains of pigment which have been alluded to were abundant both intercellularly and within phagocytes. One or two granulomas were found in the spinal cord itself; they consisted almost exclusively of endothelioid cells. Torula cells were not observed in them.

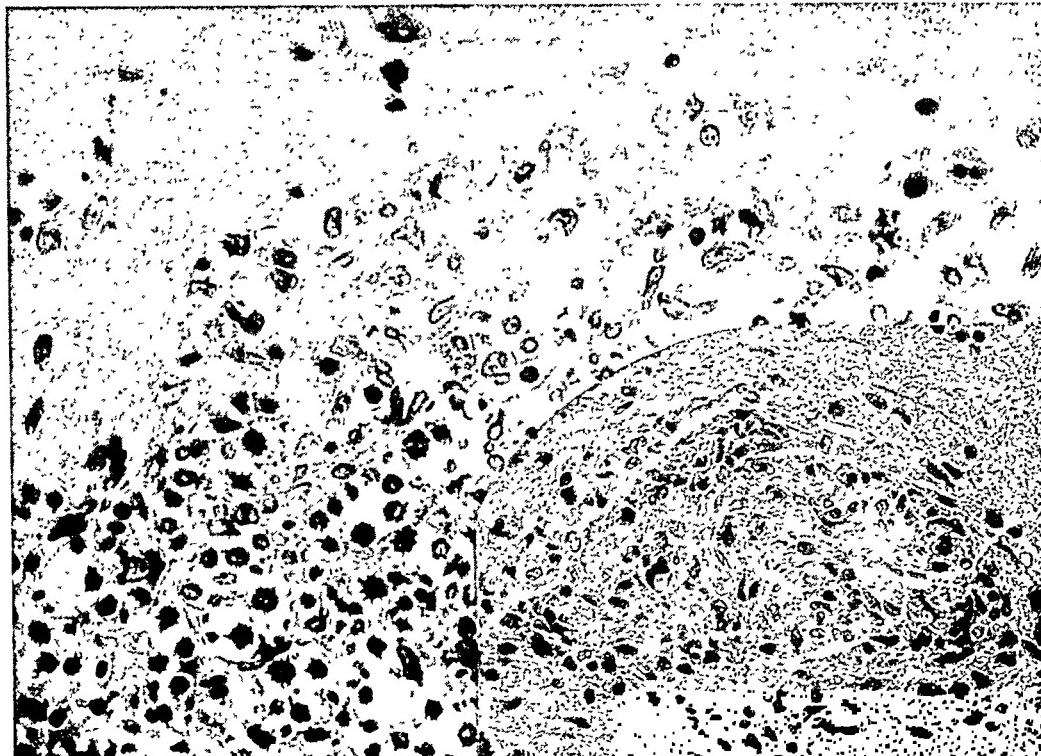


Fig. 4.—Wall of a cerebral cyst. The black spiculated bodies are yeast cells, which were distorted by the fixative. This field was selected to show endothelioid reaction rarely observed in the wall of a cyst. The insert shows a section of one of the granulomas rarely found in the spinal cord.

*Summary of Histologic Observations.*—*A. Torular Phase:* Yeast cells were most abundant in the meninges of the cerebrum, cerebellum and spinal cord, but cysts of the brain as well as the glomeruli and tubules of the kidneys also contained considerable numbers. They occurred sparingly in the pancreas and spleen. Whether the extensive chronic diffuse granulomatous changes in the latter might indicate that yeast cells were present at a previous time but had been destroyed or otherwise removed is a matter of speculation and will be discussed later in the paragraphs on Hodgkin's disease. They were not observed in the lungs or lymph nodes. Their absence in the latter is difficult to explain

in view of the fact that scrapings from lymph nodes had revealed them at necropsy and that they were present in tissue removed for biopsy.

As to the type of tissue reaction, it was consistent with that for *Torula histolytica* infection in general, i. e., either minimal or nil. In the brain substance and pancreas it was entirely absent. In the kidney it occurred in a mild, subacute form around some of the glomerular tufts and in the interstitial substance. In the cerebrospinal meninges there were a scattering of lymphoid cells and in places extensive hyperplasia of large, clear cells with rarefied cytoplasms, which were somewhat reminiscent of xanthoma cells. They were probably endothelial macrophages. In short, the severity and kind of tissue reaction varied from organ to organ.

*B. Hodgkin's Disease:* The major histologic problem of the case centered around the tissue reaction in the spleen and the lymph nodes, including the mediastinal mass.<sup>3</sup> In all sections there were (1) more or less loss of architecture and (2) reticulo-endothelial hyperplasia, with the presence of Dorothy Reed cells. The peribronchial lymph node exhibited such a degree of fibrosis that there could be no hesitation in diagnosing the changes as those of Hodgkin's disease; at least they were compatible with that disease. For the most part, however, the endothelial changes far overshadowed the lymphocytic and fibrous tissue changes, which for a long time delayed a final diagnosis of Hodgkin's disease. In principle, the reaction was an unusually massive, chronic diffuse, hyperplastic lymphadenitis, but peculiar in the same direction as obtains in Hodgkin's disease. Thus, it is admitted that such changes could still be produced by various agents, i. e., whether the process was torular in etiology or whether the conventional Hodgkin's disease was its basis, the end-result could be the same histologically. At this time we cannot decide finally which mechanism obtained in our patient, but it is at least permissible to submit a proposition in the same sense that the geometrician would open a problem for solution, namely, that *T. histolytica* is one of perhaps several micro-organisms which may evoke the histologic picture of Hodgkin's disease. The actual solution of the problem remains to be accomplished.

Comparing the histologic changes in the spleen, lymph nodes and mediastinal mass, successive steps can be traced between the obviously torular processes in the spleen and the processes definitely due to Hodgkin's disease in the lymph nodes and mediastinal mass. In the spleen, torula cells themselves (sometimes surrounded by granulomas) were seen; the grade of endothelial hyperplasia was mild, although there

3. Histologic sections were filed in the Army Medical Museum, Washington, D. C., (their number 36461) and in the Laboratory of Dermatological Research, University of Pennsylvania (accession number 2249).

was marked fibrosis. In the lymph nodes, in which the torula cells were absent, the endothelial cells were highly hyperplastic, and Dorothy Reed cells were observed. Fibrosis was present here also. In the mediastinal mass, endothelial features dominated; there were large numbers of Dorothy Reed cells, and, furthermore, the large size of the endothelial nuclei and the peripheral infiltrative characters indicated a high order of proliferative changes bordering on neoplasia. The interpretation placed on these observations is that the reticulo-endotheliosis was less active in the spleen, more highly developed in the lymph nodes and of severest grade—almost neoplastic—in the mediastinal mass. This is consistent with the gross anatomic findings.

*Reconciliation of Torula Cells with Features of Hodgkin's Disease.*—Such a reconciliation cannot be made readily in view of what has just been stated about (1) the presence of torula cells in a spleen that was far from the type seen in Hodgkin's disease histologically and (2) their absence, at least histologically, in the highly proliferative mediastinal mass which conformed more to the type seen in Hodgkin's disease. However, final decision should be reserved on this point when it is recalled that torular colonies were secured in cultures from the mediastinal mass. The possibility must be kept in mind that there are ultra-microscopic forms of Torula.

#### STUDIES OF CULTURES

The culture from our case was one of twenty strains of cerebrospinal Torulae which have been under systematic investigation in this laboratory over several years. Hence there was an unusually favorable opportunity for comparison with previously studied strains of cerebrospinal Torulae.

*Solid Mediums.*—On Pennsylvania medium<sup>4</sup> (a modified Sabouraud medium), colonies were not distinctive during the earlier stages of growth. They had a creamy, pasty quality which is common to monilias and several other species of yeast organisms. Growth was rapid, colonies attaining a diameter of 2 cm. within five weeks. At this time the colony was still pasty; it had a smooth, regular, moderately shiny surface, but had become creamy yellow. Within three months it had become deep cocoa brown, and the surface was nodular or lumpy. In some colonies the surface became divided (at about four weeks) into sectors, some of which became yet deeper brown with age and within which the surface was smooth instead of lumpy. This was an expression of dissociation—a feature which occurs in several other strains of cerebrospinal torulas. At times the colony became so soft as to flow to the bottom of the tube, which is highly characteristic for *T. histolytica*. However, Dr. Fitchett's strain was one of three in our series of twenty which tended to remain solid, as shown in figure 5A. The fluid colony (fig. 5C) was the more unusual. The medium underlying the colonies became turbid and brown. On bromphenol blue agar ( $p_{H}$  4.6) the center of the colony was deep blue, while only the margins were brown.

4. Weidman, F. D., and Spring, Dorothy: Arch. Dermat. & Syph. 18:829, 1928.

*Fermentation Tests.*—The material used as inoculum was tested for purity by both plating and direct examination of stained smears. Cultures from 4 to 5 days old, growing on Pennsylvania medium, served as the source of the inoculum. The test medium was the broth recommended by Castellani<sup>5</sup> in his extensive experiments on bronchial moniliasis. Bromcresol purple served as the indicator. The dulcite and arabinose broths were sterilized by passage through Berkefeld filters; the remaining fermentable substances were tristerilized in the Arnold apparatus. The material used in the control experiments consisted in a known highly active fermentor in the form of yeast, and in uninoculated tubes of the various solutions of sugar, which served as standard for comparison of

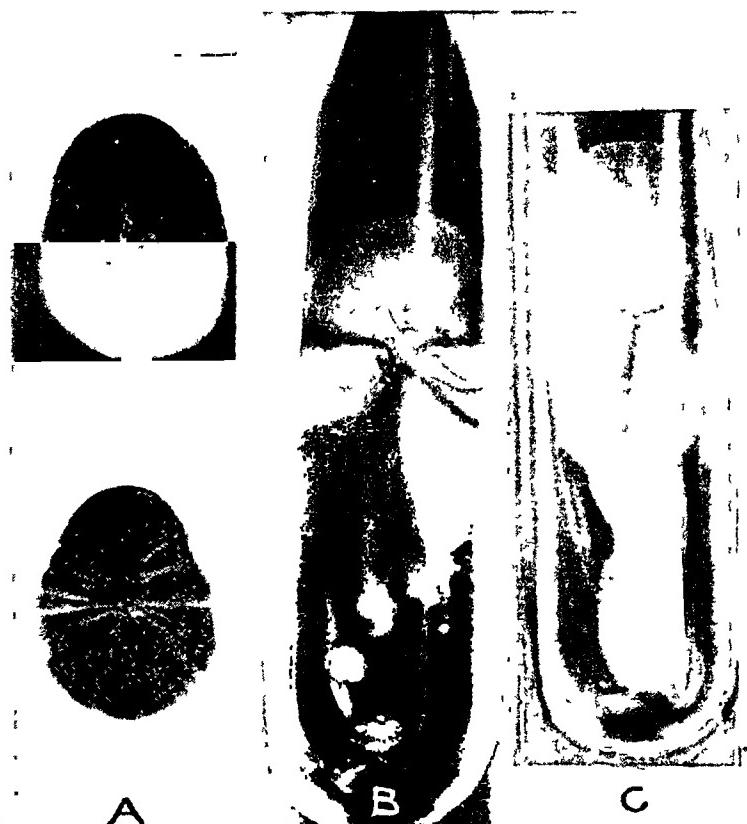


Fig. 5.—A, colony 1 month old (Pennsylvania medium); the elevated parts are expressions of dissociation. B, colony 2 months old; the darker (dissociated) sectors were pale chocolate brown. C, colony 10 weeks old; this is an example of the watery type of colony, with dripping to the bottom of the tube.

color. An additional control measure consisted in inoculations into tubes containing indicators but from which the sugar was omitted; this served as a test

5. Personal communication from Dr. Castellani. Meat extract is not to be employed on account of the small percentage of dextrose which it contains. However, 0.5 per cent sodium chloride is incorporated. Formula: To peptone water (peptone 1 per cent, sodium chloride 0.5 per cent) add 1 per cent of the desired sugar. This medium is approximately 1.5 per cent acid, and enough sodium hydroxide should be added to make it neutral ( $p_{H}$  7). Castellani used litmus extract as an indicator for the production of acidity.

against fermentable constituents in the broth itself. The substances tested were: dextrose, maltose, dextrin, saccharose, galactose, mannite, lactose, salicin, dulcite, melitose, arabinose, adonitol, inulin, levulose and sorbite.

Within six days the production of acid by our strain was perceptible, and within nine days the acid reaction was strong, but only dextrose, levulose and saccharose were thus fermented. Readings were continued until the nineteenth day. Gas was not produced under our conditions (diffuse daylight at room temperature).

Compared with the entire series of twenty strains, our strain agreed in that not one culture produced gas. It agreed with three other strains as to the production of acid.<sup>6</sup> Incidentally, for the group as a whole it was found that there was considerable variation in the production of acid. At least dextrose and levulose were fermented by all twenty strains. To illustrate: Only dextrose and levulose were fermented by seven of the strains; dextrose, levulose and saccharose were fermented by four strains; melitose, dextrose, levulose and saccharose were fermented by four strains. Inulin, dextrose, levulose, saccharose and melitose were fermented by two strains, and these five sugars and mannite by two strains. *T. histolytica* is indeed a weak fermentor.

*Microscopic Appearance in Culture*.—In material from a 4 month old colony (the characteristics were more definite in old cultures), the features were those of a cryptococcus. Thus, hyphae were not produced, and asci were not demonstrable either after a sojourn of cells on plaster of paris blocks or by staining by Beauverie's technic.<sup>7</sup> The cells were fairly large and spherical or somewhat pyriform and had thick shells. Budding occurred only occasionally in these older cultures. Younger cells contained smaller or larger granules, while larger ones either had a solid hyaloid interior or contained coarse granules or spherules which stained red with sudan III. The latter were not sufficiently uniform in size or consistent in number (4 or multiples of 4) to indicate that they represented asci. The mucinoid envelop, which is characteristic of *T. histolytica* was readily demonstrated around many of the larger cells by the wet india ink technic (a solid particle of culture is stirred into a minute drop of india ink; the cover slip is applied immediately, and the particle is examined under the microscope). As compared with the other nineteen strains of the series studied, our strain had a particularly thick capsule in the sediment in fermentation tubes. The capsule was even thicker in the mediums containing inulin and adonitol. It was moderately thick in all the other sugars tested, except lactose and arabinose. In the latter two the capsule could not be identified.

*Mycelium Formation*.—While our strain was essentially nonmycelial, hyphae were occasionally observed in very old colonies (four months old). Such hyphae really represented highly elongated yeast cells, which, however, did not branch. They had a double contour. The interior contained small numbers of smaller and larger hyaloid spherules lying in a clear, colorless matrix. The terminus of such a cell frequently consisted of a series of coalescent knobby arrangements, each of which was reminiscent of one of the spherical cells but had not as yet

6. Analysis of the protocols of the cases represented by the three strains in respect to symptoms and morbid anatomy disclosed that the case of Freeman and Weidman fell into this fermentation group. The changes in that case resembling those in Hodgkin's disease will be discussed later.

7. Guillermond, A.: *The Yeasts*, translated by F. W. Tanner, New York, John Wiley & Sons, Inc., 1920, p. 52.

become abstricted from the elongated one. Freeman and Weidman<sup>8</sup> witnessed such formations on a previous occasion, but only in fluid medium which was very old, whereas with our strain it was also observed on solid medium. In any event, such pseudomycelium formation is to be regarded as exceptional and does not militate against classifying the organism as *Torula*.

In short, all the characteristics—gross, microscopic and biologic—were compatible with those which have already been postulated for *T. histolytica*, as understood at present. We have no hesitation in identifying the organism as such.

#### INOCULATION OF ANIMALS

Two cats, one white rat and one monkey were used. All the injections were made intrameningeally, except in the monkey, which was inoculated subcutaneously. A hole was made at the summit of the calvarium anteriorly with a small drill; the lesion was allowed to heal, and thereafter (from seven to ten days) injection was made by a hypodermic needle through the bony defect. In all cases an incision was made over the bone to secure a clear field, making certain that the material entered the meninges or at least the cranial cavity.

*Cats.*—Cat 1 received a freshly isolated culture from a rat which had been experimentally infected. Four and one-half weeks after injection it was killed, but examination gave negative results for infection with *Torula*. The second cat, which was inoculated with a culture which had not been (supposedly) increased in virulence by passage through a rat died within nine days. Small numbers of torula cells were observed in the washings of the meninges, but there were no recognizable torular lesions in the viscera. At most, the kidneys were large and white and had granular surfaces, suggesting chronic parenchymatous nephritis. There were a few small, turbid, gray markings in the liver suggesting focal necrosis. The solitary follicles in the spleen were enlarged.

Microscopically, the liver and kidneys were free from torular lesions. In the brain, however, there was marked thickening of the meninges as the result of round cell (largely plasma cell) infiltration. This extended well downward into the sulci, but there were no cysts present such as were seen in the human subject. There were torula cells in large numbers, some occurring in chains. The mucinoid envelop by which they were surrounded was not so heavy as is commonly observed. A further departure from the usual observations was the presence of numerous pink granules on the exterior of the yeast cell, such as are often seen on the conidia of aspergilli and certain other species of fungus. In short, the tissue reaction in this cat varied definitely from that seen in other experimental animals, first, in the scarcity of mucin around the yeast cells and, second, in the type of reactive cells. It appears that the infection managed to barely maintain a foothold and progressed but slowly, and that the parasite was growing in a vegetative rather than a parasitic way.

*Rat.*—The rat died sixteen days after inoculation. Lesions could not be identified at necropsy. Microscopically, the most severe involvement was observed in the meninges, which were thickened by the presence of torula cells. Lymphocytes were not numerous and were confined to certain accumulations around the larger blood vessels. Yeast cells occurred in such masses as to make deep indentations in the cerebral cortex, besides occurring as comparatively large cysts within

8. Freeman, W., and Weidman, F. D.: Arch. Neurol. & Psychiat. 9:589, 1923.

its substance. There was no reaction around the cysts. In the pancreas there were several smaller or larger foci of torula cells. Some were large enough to be classified as small cysts. Most of them were not attended by a cellular reaction in the periphery; however, other foci were surrounded by a dense layer of lymphocytes. Giant cells and endothelioid cells were absent. The spleen, likewise, was affected, but only through the presence of three or four isolated individual cells, which were seen only after careful search through an entire histologic section. In the kidney there were a few clusters of torula cells in certain of the glomeruli. In neither of these organs was there any inflammatory reaction around the foci of yeast cells. Torula cells were not found in the liver. There was thus a definite parallelism between the infection in man and in the rat, as to both the

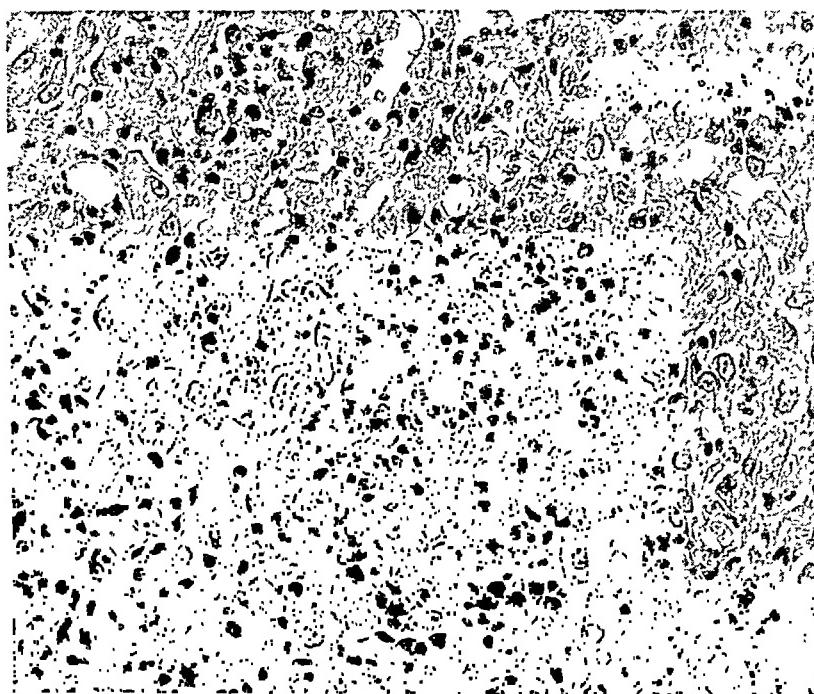


Fig. 6.—Nodule in scalp of an experimental cat; the circular space contains a budding torula cell.

organ involved and the extent of the involvement. The changes resembling those in Hodgkin's disease, however, were not reproduced.

*Monkey*.—A *Macacus rhesus* was inoculated intradermally in the shoulder and the groin. The tests were confined to the skin in this animal, because they were incidental to studies of cutaneous torulosis in man, in which it was sought to learn whether the skin could serve as a portal of entry and whether intradermal injection would be followed by generalized infection. Our strain was the one selected for inoculation. Within a week a nodule 2 cm. in diameter had developed, which was soft like an abscess but which did not exhibit any surrounding congestion. The summit of the nodule sloughed, revealing an underlying core; small quantities of pus exuded, which contained enormous numbers of torula cells. The entire lesion regressed spontaneously within two weeks, leaving a dense scar. The animal died of enteritis two months after inoculation, but did not exhibit any torular lesions of the viscera or brain at necropsy. In short, whereas frank lesions developed in

the skin, metastasis did not take place to the internal organs. The details of these experiments will be found recorded by one of us<sup>9</sup> in another place.

*Endotheliosis in Experimental Animals Resembling That in Hodgkin's Disease.*—In connection with this series of twenty strains of Torula previously cited, intrameningeal inoculations were made over a period of several years into cats, white rats and mice. The spleen and lymph nodes of all these animals so far as these were available, were restudied by us with special reference to changes resembling those in Hodgkin's disease. Unfortunately, the lymph nodes had been preserved for microscopic examination in only a few cases, because only the torula cell entered into consideration at that time. There thus remained for the present study the following materials: thirteen spleens and three lymph nodes of rats and four spleens and three lymph nodes of cats.

In only one animal, a cat, was anything observed which approached the changes resembling those of Hodgkin's disease in our patient. Thus, immediately anterior to the primary site of inoculation on the scalp was a nodule 2 mm. in diameter. Histologically, it consisted of indefinitely outlined, more or less stellate cells richly disposed on a delicate reticulum; i. e., the histologic picture was precisely that seen in the mediastinal mass of our patient, with the presence, in addition, of rather numerous torula cells. In the rats there was a definite tendency toward fibrillar fibrosis in the reticulum of the spleen, together with increase of the macrocytes, which are normal in the spleen of that animal. There was also definite hyperplasia of certain large splenocytes, which occurred in small numbers around the splenic nodules of the rats. In general, however, there were no definite changes of Hodgkin's disease in any of the animals, except in the cat first mentioned. The changes in this animal, however, were such as cannot be reconciled with those of any previously described torulosis. At the same time, they were so distinctive and reproduced so faithfully the histologic picture in the mediastinal mass of our human subject that they must have some relation to the torular processes. It should be recalled, however, that none of the rats lived longer than seven weeks after inoculation—entirely too brief a period within which definite changes of Hodgkin's disease should be expected to occur.

#### COMMENT

In Freeman's<sup>1</sup> extensive monograph only three instances of multiple visceral involvement are described. In our patient the kidneys were severely involved, and the pancreas, spleen and lymph nodes were definitely affected, to say nothing of the cerebrospinal involvement. Only the lungs (the liver was not available for histologic examination) were unaffected. Incidentally, the renal involvement brings attention to the urine as an additional source of identification of torulosis; according to Freeman's monograph, the yeast cell was found in urine once—in his case 5. This was also a case with extremely wide involvement of the viscera, particularly of the lymph nodes. There can be little doubt that there was invasion of the blood stream in our patient; this was proved by the presence of torula cells in renal blood vessels; yet the blood culture was negative.

*Relationship to Hodgkin's Disease.*—Hodgkin's disease, to say nothing of tuberculosis, has already been touched on in the literature of torular infection. It would seem to be more than a coincidence that a condition like Hodgkin's disease, which is excessively rare like torulosis, is the one to receive mention, to say nothing of the suggestive relation which we observed in our patient. Indeed, it is cited specifically in the report of two of the forty-four instances collected by Freeman (that of Freeman and Weidman and that of Smith and Crawford). Thus,

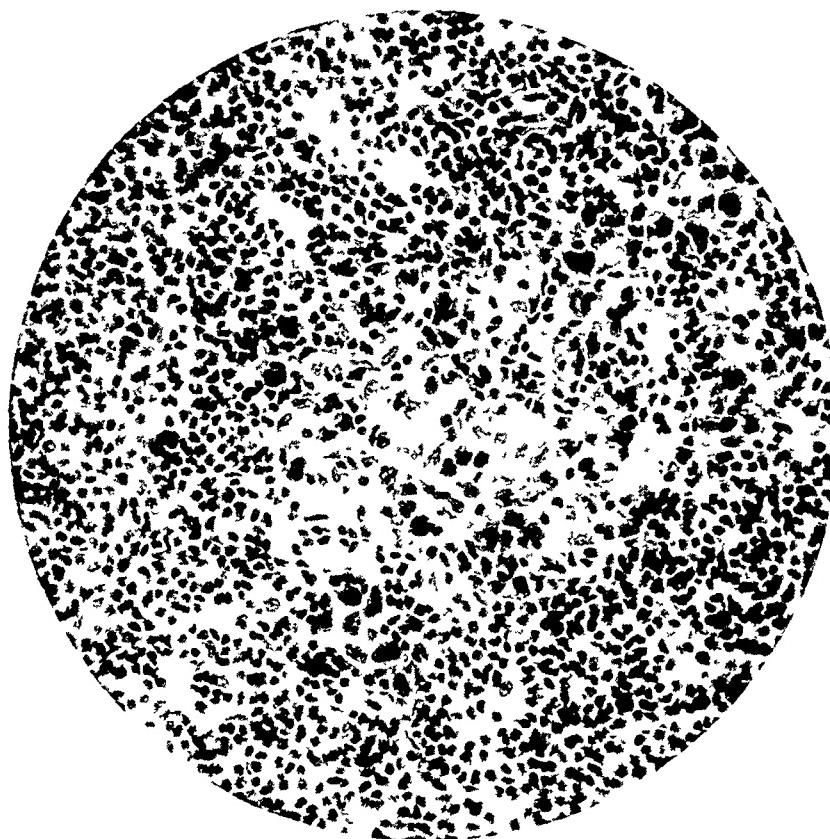


Fig. 7.—Section of mesenteric lymph nodes in Freeman and Weidman's case, showing a localized focus with Dorothy Reed cells.

in Freeman and Weidman's case there were enlarged nodes in the left axilla and about the head of the pancreas, the largest measuring 9 by 5 by 2.5 cm. Furthermore, biopsy performed five years previously had shown Hodgkin's disease, and the nodes had diminished under irradiation therapy. However, the nodes examined at necropsy were not quite typical of Hodgkin's disease, although they were highly suggestive, and the determination of a specific infectious factor (*Torula*) assisted in dismissing serious considerations of Hodgkin's disease. No yeast organisms were found in sections of lymph nodes secured at necropsy, but when used for the inoculation of guinea-pigs they were recovered

from the nodes at the head of the pancreas. It should be recorded, at least, that the strain from this subject fermented dextrose, levulose and saccharose, i. e., the same group that was fermented by our own strain. Since two other strains among the twenty which we studied also fell into this group, special significance cannot be invoked, at least not at present.

Although there was no enlargement of lymph nodes in Smith and Crawford's case, there was swelling over the left scapula, which was regarded histologically as atypical of Hodgkin's disease. This lesion was examined seventeen months before necropsy. After necropsy, when the diagnosis of torulosis had been made, reexamination of the sections disclosed the presence of *Torula*.

Enlargement of lymph nodes due to a nontorular agent, i. e., the tubercle bacillus, has occurred frequently in torulosis, without provoking thoughts particularly of Hodgkin's disease. In one such case there was cervical adenopathy (Türk), and in a second the lymph nodes were removed surgically a number of years previously (in Evans' second case). Enlargement of mediastinal nodes and "acute inflamed" mesenteric nodes were found in Williams'<sup>10</sup> patient, but were not examined microscopically. Furthermore, in the patient studied by Rusk and Farnell there was an abscess of the axilla which was very slow in healing. A chronic axillary abscess also occurred in Wildman's case and in Rusk and Farnell's second case. Further details of the foregoing cases will be found in Freeman's monograph.

In both the cases just cited it may be noticed that the diagnosis of Hodgkin's disease was made with decided reservations. We reexamined the original sections in the case of Freeman and Weidman, searching particularly for torula cells which might have been overlooked, but must leave the situation as originally stated; that is, the histologic changes were suggestive, but were not definitely those of Hodgkin's disease. As there were not sufficient clinical symptoms, such as anemia and pronounced adenopathy, to support the histopathologic changes, we feel constrained to exclude all these cases as Hodgkin's disease and to surmise that they were but instances of general, low grade lymphadenitis such as are often classified simply as adenopathies.

But at such a juncture consideration of our own case enters to give a special and renewed significance, because the symptoms were so definitely those of Hodgkin's disease that there was no hesitation in making that diagnosis clinically. Thus, the emaciation bespoke extreme weakness, and there were definite anemia and marked adenopathy. The question at issue is whether the torulosis was responsible for the symp-

10. Williams, J. R.: M. J. Australia 2:185, 1922.

toms of Hodgkin's disease, i. e., whether it is one of several organisms or noxae which may induce such symptoms, or whether the torulosis was superimposed on preceding Hodgkin's disease. The occurrence of torula cells in the lymph nodes, in both the specimen secured at biopsy and the specimens obtained at necropsy throws additional suspicion on torulosis as the etiologic agent, but without quite incriminating it. (It may be recalled that *Torula* was also cultured from lymph nodes at the head of the pancreas in Weidman and Freeman's case.) It can be explained just as reasonably that with such extensive systemic involvement there was obviously an infection of the blood stream, and that lymph nodes should be expected to participate equally with the kidney, pancreas and other organs.

One cannot dismiss the association of these two diseases summarily, in view of both the unsettled status of the causation of Hodgkin's disease (also its scope and limitations, such as Hodgkin's sarcoma) and the possibility that peculiarities in the constitution of persons or of their lymph glands might permit the development of symptoms of Hodgkin's disease from a torular lesion as well as from infection due to the tubercle bacillus, the pseudodiphtheria bacillus and other micro-organisms which have from time to time been advanced as etiologic agents in Hodgkin's disease. In any event, the torular reaction might well serve as a tool in the investigation of the etiology of Hodgkin's disease as revealed histologically. Similarly, since the cells do not always show up well in routine hematoxylin and eosin sections, special precautions should be taken to attempt to demonstrate them in the brain in future cases of Hodgkin's disease, at least if there are neurologic symptoms.

*Type of Tissue Reaction Against Torula Cells.*—This conformed with that of other cases of infection with *T. histolytica*; i. e., it was either absent or scanty in the brain substance and generally scanty but sometimes tuberculoid in the other organs.

*Portal of Entry.*—This topic merits special attention here only in case it is concluded that the yeast cells were the cause of the generalized lymphadenopathy. In that case the intestines would appear to have been the primary portal of entry instead of the lungs and the nasopharyngeal passages, as is usually the case in torulosis. Such a portal is entirely consistent with the biology of yeast cells; this is illustrated by the fact that Tanaka<sup>11</sup> observed 10 per cent *Saccharomyces* in apparently normal mesenteric lymph nodes examined at necropsy.

*Pathogenicity of the Micro-Organism.*—This particular strain conformed to others of *T. histolytica* in being pathogenic for the white rat but not for the cat. It was also pathogenic for the skin of a monkey.

11. Tanaka: J. Path. & Bact. 23:350, 1920.

## SUMMARY

The fourth case of widespread, generalized torulosis, which is usually confined to the cerebrospinal nervous system, is reported. It occurred in a Negro, 18 years of age, who had had symptoms of Hodgkin's disease for three years. In forty-four collected instances there have been two additional cases in which the histologic picture of Hodgkin's disease was also approached, but only in our own case has the full symptomatology (weakness, anemia and massive adenopathy) of Hodgkin's disease been recorded. Nevertheless, it would be premature to regard torular infection as one of the causative agents of Hodgkin's disease, although changes were highly suggestive of such an etiology. The presence of extensive renal involvement calls attention to the necessity for examination of the urine for torula cells as they have been observed in the urine at least once. Attention is called anew to the possibility that the intestines may serve as the primary portal of entry for torulosis. Cases of Hodgkin's disease with meningeal symptoms demand examination for torular infection.

# INFLUENCE OF VARIOUS DIETS ON EXPERIMENTAL AMYLOIDOSIS IN MICE

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Amyloidosis is found only infrequently at autopsy in China, although chronic tuberculosis, osteomyelitis and other kinds of chronic suppurative processes are more common than in the western countries. Kuczynski<sup>1</sup> reported the infrequency of amyloidosis also in the Japanese. Tanaka<sup>2</sup> stated that no advanced cases of amyloidosis have been observed in Japan but that in cases of chronic suppuration the occurrence of microscopic deposits of amyloid is frequent. He attributed the milder degree of amyloidosis to the low protein content of the diet of the Chinese and Japanese. Kuczynski<sup>1</sup> produced amyloidosis in mice not only by parenteral injection of nutrose but also by feeding them certain proteins such as nutrose and cheese. He believes that a protein diet favors the production of amyloid. This has been disputed by Jaffé.<sup>3</sup> According to him the amyloidosis which develops in mice fed on cheese, as in Kuczynski's experiments, is caused by the enteritis which may develop and is not due directly to the cheese. From Jaffé's experiments, it appears that the protein diet as well as cholesterol and fat can delay the development of experimental amyloidosis in mice. According to Grayzel<sup>4</sup> and his collaborators, mice fed on liver preparations are not susceptible to amyloid degeneration. These discrepancies and the fact that in autopsy material in China and Japan severe amyloidosis appears to be uncommon led us to undertake the following experiments.

## MATERIAL AND METHODS

*Series A.*—White male mice were used. The exact age was not known, but all weighed between 25 and 30 Gm. at the beginning of the experiment. The animals were divided into four groups. One group was fed on a diet containing a large

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1. Kuczynski, M. H.: *Klin. Wchnschr.* **2**:727, 1923.
2. Tanaka, Y.: *München. med. Wchnschr.* **57**:1383, 1910.
3. Jaffé, R. H.: *Arch. Path.* **2**:149, 1926.
4. Grayzel, H. G.; Jacobi, M.; Maslow, H., and Warshall, H. B.: *Proc. Soc. Exper. Biol. & Med.* **28**:172, 1930.

amount of animal protein and just enough other substances to sustain life. The composition of the diet was as follows: meat scrap powder,<sup>5</sup> 91 parts; brewer's yeast, 5 parts; cod liver oil, 2 parts, and agar-agar, 2 parts.

The second group of mice was fed on a diet containing a large amount of plant proteins and the necessary quantity of adjuvant substances to make the diet adequate to sustain life. It was composed of 91 parts of soy bean flour,<sup>6</sup> 4 parts of brewer's yeast, 1 part of cod liver oil and 4 parts of the following mixture:

	Gm. or Cc.
Sodium chloride (NaCl).....	0.173
Magnesium sulphate (anhydrous) (MgSO <sub>4</sub> ).....	0.266
Monosodium phosphate (NaH <sub>2</sub> PO <sub>4</sub> .H <sub>2</sub> O).....	0.347
Dipotassium phosphate (K <sub>2</sub> HPO <sub>4</sub> ).....	0.954
Monocalcium phosphate (CaH <sub>4</sub> (PO <sub>4</sub> ) <sub>2</sub> .H <sub>2</sub> O).....	0.540
Ferrous citrate .....	0.118
Calcium lactate .....	1.300

The third group of mice were given a diet consisting chiefly of carbohydrate. Its composition was as follows corn-starch, 73 parts; soy bean flour, 15 parts; cod liver oil, 2 parts; brewer's yeast, 5 parts, and the aforementioned mixture, 5 parts.

The fourth group was fed on a normal stock diet consisting of: ground corn, 600 Gm.; cracked wheat, 300 Gm.; dried milk, 60 Gm.; meat scrap, 90 Gm.; sodium chloride, 10 Gm., and lettuce, hard bread and milk.

Water was given *ad libitum* to the four groups. The mice were fed on these diets for at least thirty-five days preceding the first injection in order to adapt them to the new diets. Then they were given daily 0.5 cc. of 3 per cent sterile solution of nutrose in physiologic solution of sodium chloride by intramuscular injection into both hindlegs. All the mice, other than those which died spontaneously or were killed when in moribund condition during the early period of the experiment, were killed by inhalation of chloroform after receiving thirty daily injections. Cultures were taken from the muscles of the hindlegs of all the mice which died before the completion of the series and also from the majority of those which were killed. Since the spleen, liver and kidneys are considered by all authors as the most common and constant sites of amyloid deposition, portions of these three organs were fixed in 10 per cent solution of formaldehyde and in 80 per cent alcohol. Paraffin sections were stained with hematoxylin and eosin and with congo red.

*Series B.*—Another group of similar experiments was carried out with the following differences in technic: Fifty daily injections were given instead of thirty. Animals which did not receive injections were fed the diets for eighty days to see whether spontaneous amyloidosis developed. A small number of animals was fed on the stock diets for thirty days and then given fifty daily intramuscular injections of sterile saline solution. In this series of animals the sites of injection were not examined bacteriologically for possible infection, but all the animals found to be suffering from gross infection were discarded.

5. The meat scrap powder contains 50 per cent protein. No salt mixture was added to this material because of the high inorganic salt content found on analysis, which showed: total chlorides (calculated as sodium chloride), 0.96 per cent; calcium (high ground bone content), 11.0 per cent, and phosphorus, 4.3 per cent. These figures give the percentage by weight.

6. The percentage of protein in soy bean flour is from 41 to 43 per cent.

Sections of liver, spleen and kidney from the latter experimental animals were fixed in 80 per cent alcohol and in Zenker's fluid to which a dilute solution of formaldehyde had been added. Methyl violet and Mallory's aniline blue stains were employed.

The results are best illustrated by the accompanying tables.

TABLE 1.—*Series A, Animals Which Received Injections of Nutrose*

Diet	Number of Daily Injections	Number of Animals Receiving Injections	Number of Animals Showing Deposits of Amyloid
Meat scrap.....	30	14	6
Soy bean.....	30	12	5
Carbohydrate.....	30	16	2

TABLE 2.—*Series B, Animals Which Received Injections of Nutrose*

Diet	Number of Daily Injections	Number of Animals Receiving Injections	Number of Animals Showing Deposits of Amyloid
Normal.....	50	16	9
Meat scrap.....	50	13	6
Soy bean.....	50	26	7
Carbohydrate.....	50	18	0

TABLE 3.—*Animals Receiving Injections of Saline Solution*

Diet	Number of Daily Injections	Number of Animals Receiving Injections	Number of Animals Showing Deposits of Amyloid
Normal.....	50	6	2
Meat scrap.....	50	8	0
Soy bean.....	50	5	1
Carbohydrate.....	50	6	2

TABLE 4.—*Animals Which Did Not Receive Injections*

Diet	Number of Animals in Group	Number of Animals Showing Deposits of Amyloid
Normal.....	10	0
Meat scrap.....	11	0
Soy bean.....	12	0
Carbohydrate.....	11	0

TABLE 5.—*Combined Results of Series A and B, Animals Receiving Injections of Nutrose*

Diet	Total Animals in Experiment	Number Showing Deposits of Amyloid	Number Not Showing Deposits of Amyloid	Percentage Positive
Meat scrap.....	27	12	15	44.4
Soy bean.....	33	12	26	32.1
High carbohydrate.....	34	9	32	5.8
Normal.....	16	9	7	56.0

The spleen was found to be the most constant site of amyloid; this is well known and agreed on by all authors. In animals in which the specific staining of amyloid was at first doubtful, restains were performed (with controls) on the spleen with methyl violet and, when positive, the results were interpreted as positive in the other tissues and vice versa.

Histologically the pictures were similar to those described and illustrated by Jaffé.<sup>7</sup> In addition, a considerable number of giant cells which showed rose-colored amorphous material when stained with methyl violet were seen in the spleen. This picture corresponds to the "precursor" stage of amyloid as described by Grayzel.<sup>8</sup> Such evidence of amyloidosis alone was not considered positive for our diagnosis, and only in cases in which the amounts were unequivocal was a positive diagnosis for amyloid made.

#### COMMENT

The greatest percentage of mice showing amyloidosis occurred in the group fed on normal diets, while the group fed on a high carbohydrate diet showed the smallest percentage. The severity of the change was not appreciably greater in any specific group.

The percentage of mice showing amyloidosis was strikingly low in the group fed on the high carbohydrate diet. The difference is one of frequency of occurrence rather than of degree of change in the individual mice of the different groups. It is evident that mice on normal, meat and soy bean diets, all containing protein, were susceptible to amyloidosis. The results do not contradict Kuczynski's<sup>9</sup> assumption that a protein diet favors the experimental production of amyloidosis in mice.

Kuczynski,<sup>9</sup> using cheese and egg albumin, and Smetana,<sup>10</sup> using Swiss cheese, found that the high protein diet favored the production of amyloidosis in mice, while Jaffé<sup>3</sup> found that beef heart powder not only delayed the formation of amyloid but even "protected" mice against it. According to Grayzel<sup>4</sup> and his collaborators, liver powder had an effect similar to that of beef heart powder and delayed the production of amyloid in mice. Grayzel<sup>8</sup> also stated that resorption is accelerated by liver once the injections are discontinued, provided advanced amyloidosis has not set in. It appears that not all proteins have the same effect in influencing the production of amyloid in mice. Furthermore, the standard laboratory diet—consisting of white bread,

7. Jaffé, R. H.: Arch. Path. **1**:25, 1926.

8. Grayzel, H. G.; Jacobi, M.; Warshall, H. B.; Bogin, M., and Bolker, H.: Arch. Path. **17**:50, 1934.

9. Kuczynski, M. H.: Virchows Arch. f. path. Anat. **239**:185, 1922.

10. Smetana, H.: Bull. Johns Hopkins Hosp. **37**:383, 1925.

oats and skimmed milk—in Jaffé's experiments—as well as Grayzel's<sup>8</sup> standard diet favored amyloidosis in mice, whereas we observed that the mice fed chiefly on corn-starch were not likely to show the change. Whether corn-starch exerts a protecting effect on experimental amyloidosis in mice as produced by injections of nutrose or whether the effect is due to the failure to ingest sufficient amounts of the substances which may be necessary for the formation or deposition of amyloid is not determined. Therefore, the influence of different kinds of carbohydrate may also differ. It is probable that the kind of protein or of carbohydrate favors the formation of, or protects against, amyloid, respectively. Even if Kuczynski's theory that a diet high in protein favors experimental amyloidosis were accepted fully, our results could hardly be applied to human pathology. Although the Chinese eat soy bean in many forms and in considerable quantity, cases of amyloidosis are seldom observed in China.

Although many investigators have used Kuczynski's<sup>1</sup> method of producing amyloidosis in mice, no critical application to human pathology is found in the literature. Nutrose is definitely accepted by all authors as a substance efficient in inducing experimental amyloidosis, yet there is no absolute certainty in that respect. According to Letterer<sup>11</sup> it is not possible to produce amyloid in every case by parenteral administration of nutrose, but Smetana<sup>10</sup> claimed almost 100 per cent positive results after thirty injections of solution of nutrose. It appears that the type of stock diet used modifies to a great extent the time and number of injections required to produce experimental amyloidosis by parenteral injections. This factor is well shown in Grayzel's<sup>8</sup> work. In his group of mice in which various combinations of vitamins were used the total incidence of amyloid, even after as many as sixty-seven injections, was strikingly low. Uchino<sup>12</sup> emphasized the added effect of bacterial infection which, according to our study, is a sufficient but not a necessary condition for the development of amyloid and may act as an adjuvant. In a number of the control animals, even in the group fed on a carbohydrate diet and receiving injections of sterile saline solution, positive amyloid reaction occurred. This may indicate that corn-starch, while it appears to exert a protecting influence on experimental amyloidosis as produced by nutrose, is not so efficient when infection is present. Although no bacteriologic examinations were made, it is assumed that the probable explanation for amyloidosis in these animals must have been infection, for no amyloidosis developed in any of the animals which did not receive injections, regardless of what diet they were fed.

11. Letterer, E.: Verhandl. d. deutsch. path. Gesellsch. **20**:301, 1925.

12. Uchino, S.: Beitr. z. path. Anat. u. z. allg. Path. **74**:405, 1925.

The exact nature of human amyloid remains obscure. In a summary review of Leupold<sup>13</sup> the differences of opinion of various investigators about this question is well discussed. No attempt at chemical analysis of the experimental amyloid appears to have been made. The supposed identity of experimental and of human amyloid has been based on the morphologic appearance. Staining reactions vary. The iodine and methyl violet stains are not always positive; this has also been observed by Smetana. Our results showed a fairly good accord between methyl violet and Mallory's aniline blue. Fairly good results were obtained with congo red.

All authors agree only on the statement that animal and human amyloid belong to the same type of protein compound. Karczag, Paunz and Németh<sup>14</sup> expressed the opinion that animal amyloid is a prephase of the human type. The mode of formation of amyloid in animals and human beings appears to be different. In animals we always observed a perifollicular deposition of amyloid in the spleen but rarely in the form of the sago spleen. Strasser<sup>15</sup> expressed a belief that in the liver of mice the formation of amyloid does not begin in the intermediate zone but usually starts in the wall of blood vessels in the interlobular spaces. The "precursor" stage described by Grayzel<sup>8</sup> cannot be evaluated at this time.

Owing to the lack of knowledge of the chemical nature of amyloid and to the inconstancy of characteristic staining properties of animal amyloid it is impossible to apply our results directly to human disease.

#### SUMMARY AND CONCLUSIONS

The influence of different kinds of diet on experimental amyloidosis produced in mice by the intramuscular injection of a solution of nutrose was investigated. Four kinds of diets were used. Three consisted mainly of meat scrap, soy bean and corn-starch, respectively (with the necessary supplement of minerals and vitamins). The fourth was the stock mixed diet for mice.

The mice fed on the normal diet showed the largest percentage of amyloidosis; the group fed on high protein diet came next, and the smallest percentage was found in the group fed on a diet rich in corn-starch. Control animals receiving injections of saline solution instead of nutrose also showed amyloid. Untreated control animals showed no amyloid.

13. Leupold, E.: *Ergebn. d. allg. Path. u path. Anat.* **21**:120, 1925-26; *Beitr. z. path. Anat. u. z. allg. Path.* **64**:347, 1918.

14. Karczag, L.; Paunz, L., and Németh, L.: *Ztschr. f. d. ges. exper. Med.* **41**:71, 1924.

15. Strasser, U.: *Ztschr. f. d. ges. exper. Med.* **36**:388, 1923.

A diet high in carbohydrate delays, protects against or fails to provide the necessary building material for the deposition of experimental amyloidosis produced in mice by injections of nutrose.

The difference between the changes in the three groups consisted more in the percentage than in the degree of severity of the changes in the individual mice.

The fact that a large percentage of mice showed positive results when fed on relatively high protein diets (both animal and plant proteins) agrees with Kuczynski's assumption that a protein diet favors the production of amyloidosis but contradicts the results of Jaffé's experiments.

Soy bean is a popular food in China. Amyloidosis is rarely found at autopsy in Chinese. The condition developed in a high percentage of mice fed on a soy bean diet and given injections of nutrose. Therefore, the low incidence of amyloidosis cannot be attributed to this feature of the Chinese diet. Even animals fed on the same diet and receiving daily injections of the same amount of nutrose exhibit individual variations of susceptibility to experimental amyloidosis. Similar variations probably exist in human amyloidosis. The inadequacy of our methods for the specific demonstration of amyloid may also play a part in the results obtained.

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# A CHEMICAL STUDY OF ARTERIOSCLEROTIC LESIONS IN THE HUMAN AORTA

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In spite of the large amount of experimental work that has been done on arteriosclerosis there are surprisingly few chemical studies of the human arteriosclerotic aorta. Gazert,<sup>1</sup> in 1899, published the first series of fat, calcium and nitrogen determinations on arteriosclerotic aortas. He showed that there was an increase of fat, ash and calcium, and a decrease of nitrogen, as the arteriosclerotic changes became more extensive. In 1906, Baldauf<sup>2</sup> again demonstrated the increase of fat, and at the same time a decrease of phosphatids with increasing calcification. Then, after Aschoff had demonstrated that a large part of the fatty material in arteriosclerotic plaques was cholesterol ester, Windaus,<sup>3</sup> in 1910, analyzed normal and atheromatous aortas for cholesterol and cholesterol esters and showed that atheromatous aortas have from six to seven times as much free, and twenty to twenty-six times as much bound, cholesterol as normal aortas. The next important work was done in 1926 by Schönheimer<sup>4</sup> who, with more modern methods, again demonstrated the increase of cholesterol and cholesterol esters. All this previous work, however, was done on extracts of whole aortas; therefore it seemed to us worth while to try to isolate, for analysis, the portions involved in intimal lesions apart from the other portions of the aorta. In this way, it was hoped to obtain a more accurate picture of the changes in the pathologic tissue alone.

## ANALYSES

The material used consisted of fresh, unfixed human aortas taken at autopsy from patients showing the usual variety of diseases found in a metropolitan hospital. The intima and part of the media were stripped away from the adventitia,

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This investigation has been aided by a grant from the Josiah Macy Jr. Foundation.

From the Department of Pathology, College of Physicians and Surgeons, Columbia University.

1. Gazert: Deutsches Arch. f. klin. Med. **62**:390, 1899.

2. Baldauf, L. K.: J. M. Research **15**:355, 1906.

3. Windaus, A.: Ztschr. f. physiol. Chem. **67**:174, 1910.

4. Schönheimer, R.: Ztschr. f. physiol. Chem. **160**:61, 1926; **177**:143, 1928.

laid with the intima down, and the media stripped off as completely as possible with a forceps and knife. The intima was then turned over, and the atherosclerotic plaques were cut out of it and classified as early, medium or late. As a control, normal intimal tissue was cut out in the same way. The early lesions were small, slightly raised, yellowish, opaque, glistening plaques, which are usually found in the arch, and may also form a streak down the posterior wall of the aorta about the orifices of the intercostal arteries. The medium lesions were thicker, yellow and gray, partly translucent plaques, containing hyalinized connective tissue and often grossly visible fat deposits. The late lesions were extensively hyalinized, calcified or ulcerated tissue.

The plaques were weighed, transferred to a paper thimble (which had been previously extracted with boiling alcohol) hung in a flask fitted with a reflux condenser and extracted continuously with boiling 95 per cent alcohol for from three to four hours. The contents of the flask were then cooled and filtered through a fat-free filter paper into a volumetric flask of appropriate size. The thimble and flask were further extracted and washed several times with ether, and the ether extracts added to the alcohol extract through the same filter, and the filter was finally washed with a little more ether. The combined extracts were then made up to volume with ether. This alcohol-ether extract was used for all the analyses. (The completeness of this extraction was checked several times by repeating the foregoing process. The weight of the residue was never more than from 1 to 2 per cent of that of the original extract.)

For the determination of the dry weight of the fatty extract, part of the original extract was dried down in a beaker, reextracted with ether, dried again, reextracted with boiling petroleum ether (boiling point from 40 to 60 C.), filtered, dried first on the steam bath and finally in a vacuum desiccator over night, and then weighed. In some cases in which the amount of fat was small, this weighed sample was redissolved in chloroform, made up to its original volume and used for the determinations of phosphorus.

*Determination of Phosphorus.*—The foregoing extract was dried down in a large pyrex test tube and ashed with perchloric and nitric acids until the solution was colorless. The process is necessarily very slow, since overheating may drive off some phosphorus. It was then transferred quantitatively to a 25 cc. volumetric flask, neutralized to phenolphthalein with concentrated sodium hydroxide, and the phosphorus determined by the method of Fiske and Subbarow.<sup>5</sup> Duplicates ordinarily checked within 5 per cent. From the phosphorus values, phospholipids were calculated as lecithin (molecular weight 810).

The cholesterol and cholesterol ester were determined on the original alcohol-ether extracts by a method devised by Schönheimer and Sperry, which will be published shortly.<sup>6</sup> Duplicates always checked within 5 per cent and ordinarily within 2 per cent. The determinations actually gave values for total and for free cholesterol. The ester fraction was obtained by difference and calculated as cholesterol oleate (in the ninth column of the accompanying table).

The cases have been arranged in groups according to the classification of the tissue as normal or as showing early, medium or late lesions, and within the separate groups the cases have been arranged according to increasing age.

5. Fiske, C. H., and Subbarow, Y.: J. Biol. Chem. **66**:375, 1925.

6. Dr. Warren Sperry of the Babies Hospital made the cholesterol determinations.

*Results of Analyses of Arteriosclerotic Plaques and Normal Tissue from*

Tissue Used for Analysis	Age, Years	Wet Weight of Tissue, Gm.	Fatty Extract, per Cent of Wet Weight of Tissue	Phospholipids, per Cent of Fatty Extract	Total Cholesterol, per Cent of Fatty Extract	Ester Cholesterol, per Cent of Total Fatty Extract	Free Cholesterol, per Cent of Total Fatty Extract
Normal tissue	12	1.46	0.87	26.1	16.2	3.01	13.2
	27	2.18	0.93	26.9	27.4	13.9	13.5
	29	2.23	1.48	26.5	27.8	13.3	14.5
	31	1.27	1.83	21.4	27.2	15.1	12.1
	60	1.88	0.90	16.3	31.2	19.4	11.8
	60	1.94	2.22	17.0	33.7	24.4	9.3
Early lesions	21-26	2.20	3.23	16.2	40.5	28.4	12.1
	27-28	2.49	3.84	19.7	41.6	31.2	10.4
	26-30	1.51	3.90	14.2	39.0	27.2	11.9
	32	1.55	4.32	21.6	40.2	22.4	17.9
	37	1.45	2.76	18.7	37.5	25.0	12.5
	37-39	2.11	6.10	15.5	48.4	32.6	15.9
	38	1.83	4.90	18.9	38.9	18.9	20.0
	42	2.06	4.55	17.5	43.6	27.7	16.0
	43†	1.58	5.32	13.1	45.2	36.9	8.3
	45	3.31	11.90	18.9	43.7	22.5	21.2
	50	2.06	3.89	16.3	42.5	31.2	11.2
	52-60	2.24	5.85	16.8	48.8	30.6	18.3
	Average.....		5.04	17.3	42.5	27.9	14.7
Medium lesions	21	1.23	3.34	12.2	44.4	32.4	12.2
	50	2.02	4.55	16.3	40.2	26.1	14.1
	52	2.45	6.20	12.2	46.6	28.3	18.4
	52†	3.75	8.08	21.6	49.5	20.7	19.8
	52	1.39	6.25	14.9	52.8	24.1	28.7
	53	2.41	8.95	18.6	50.2	26.5	23.7
	56†	2.01	9.50	18.0	54.0	37.5	16.5
	57	5.33	7.82	19.9	43.3	26.9	16.3
	57†	3.47	8.97	15.5	49.3	31.8	17.3
	57	1.75	4.97	19.1	43.6	24.2	19.0
	60	1.10	8.63	....	49.5	24.2	25.3
	66†	1.63	9.61	21.0	50.9	24.8	26.1
	Average.....		7.19	17.2	47.7	28.3	19.2
Late lesions	43†	2.59	6.55	12.4	49.4	27.0	22.4
	46-53	3.06	9.83	13.0	53.2	30.5	22.6
	49	1.18	10.85	....	46.1	22.7	23.4
	52†	4.48	8.57	18.8	52.1	31.3	20.8
	55	2.00	6.70	17.9	51.5	26.1	25.4
	56†	4.89	7.55	17.1	50.4	21.9	28.5
	57†	5.06	7.87	18.0	51.4	31.4	19.8
	59	5.77	10.90	18.3	49.6	23.1	26.5
	60	4.62	4.19	18.7	51.1	26.9	24.3
	62	2.37	8.05	16.3	52.3	19.9	32.4
	62	9.79	7.55	16.4	48.1	31.1	16.9
	66†	3.13	14.50	20.0	56.6	20.9	35.7
	66	10.78	7.93	17.9	52.8	23.5	29.2
	73	6.85	5.39	17.4	45.8	22.8	23.0
	74	12.03	9.29	19.2	54.6	24.2	30.4
	Average.....		8.38	16.9	51.0	25.5	25.4

\* In the table the averages have been calculated to show conveniently the tendencies which we have discussed. The variations in the groups are too large to give the averages exact mathematical significance.

† Cases in which both early and late or both medium and late lesions were taken from the aorta.

*Forty-Five Human Aortas to Determine Phospholipid and Cholesterol Values\**

Free Cholesterol Plus Bound Cholesterol (as Oleate), per Cent of Total Fatty Extract	Free Cholesterol- Ester Choles- terol Quotient	Diagnosis
18.5	4.00	Rheumatic heart
38.0	0.97	Rheumatic endocarditis
38.0	1.09	Generalized peritonitis (perforated ulcer)
38.8	0.80	Glomerular nephritis (hypertension)
45.9	0.61	Carcinoma of rectum (advanced arteriosclerosis)
52.3	0.38	Medial calcification of aorta
62.2	0.43	Four cases—rheumatic endocarditis; bacterial endocarditis; brain abscess and meningitis; meningitis
65.6	0.33	Adenocarcinoma of rectum, papillary cystadenoma of ovary
59.3	0.44	Chronic lymphatic leukemia, appendicitis
57.3	0.80	Endarteritis obliterans
56.5	0.50	Tuberculosis, mild arteriosclerosis
73.2	0.49	Three cases—lobular pneumonia; tuberculosis; chronic ulcerative colitis
53.3	1.06	Acute gangrenous vaginitis
64.6	0.58	Arteriolar nephrosclerosis (hypertension), slight arteriosclerosis of aorta
73.4	0.22	Astrocytoma of cerebellum
60.9	0.99	Arteriolar sclerosis, generalized arteriosclerosis
66.3	0.36	Acute bacterial endocarditis, generalized mild arteriosclerosis
72.1	0.60	Seven cases
63.9	0.56	
69.3	0.38	Rheumatic heart
60.0	0.54	Hypertension, generalized arteriosclerosis
68.4	0.65	Lobar pneumonia, medial calcification of aorta
72.2	0.67	Rheumatic endocarditis, generalized arteriosclerosis, moderate arteriolar- sclerosis
71.3	1.19	Carcinoma of colon, senile arteriosclerosis
70.2	0.89	Carcinoma of mouth
82.5	0.44	Purulent bronchitis, mild senile arteriosclerosis
63.8	0.61	Chronic nephritis and uremia, generalized arteriosclerosis, arteriolosclerosis
73.4	0.54	Carcinoma of colon, senile arteriosclerosis, hypertension
62.0	0.79	Cirrhosis of liver
67.9	1.05	Carcinoma of duodenum
70.0	1.05	Abscesses of liver, acute cholecystitis, acute cholelithiasis, medial calcifica- tion of aorta
69.3	0.73	
70.0	0.83	Astrocytoma of cerebellum
76.4	0.74	Pneumonia and appendicitis, carcinoma of stomach, mild arteriosclerosis
63.2	1.03	Carcinoma of ovary
75.8	0.66	Rheumatic endocarditis
71.6	0.97	Carcinoma of tongue, arteriosclerosis
67.1	1.30	Purulent bronchitis
75.2	0.63	Carcinoma of colon
67.4	1.15	Aplastic anemia, medial calcification of aorta
71.8	0.90	Aleukemic leukemia
67.5	1.63	Gallstones, jaundice, mild generalized senile arteriosclerosis
71.7	0.54	Adenoma of thyroid gland
72.5	1.77	Abscesses of liver, acute cholecystitis, acute cholelithiasis, medial calcifica- tion of aorta
70.6	1.24	Coronary sclerosis (advanced calcification)
62.6	1.01	Ulcer of stomach
72.9	1.25	Lobular pneumonia, generalized moderate arteriosclerosis
70.4	1.04	

## COMMENT

The chief change in the pathologic material is an increase in the amount of the fatty extract, and particularly in the proportion of cholesterol to total fatty extract. This increase apparently becomes greater with increasing severity of the lesions rather than with increasing age. The table appears to show also that as the severity of the lesions increases, there is first an increase in the percentage of cholesterol esters in the fatty extract, and then, as the process continues, a decrease, but when the ester values were plotted against the fatty extract values, it was apparent that there was too much variation to permit any curve to be drawn. A similar curve drawn for free cholesterol showed, more definitely, a slight increase in the proportion of free cholesterol with increasing severity of the lesions. However, when the ratio of free to ester cholesterol was calculated, the changes were more striking. In the normal tissue the ratio of free to ester cholesterol decreases steadily with increasing age (from 4 for the youngest to 0.38 for the oldest aorta; average, 1). In the pathologic tissue this ratio apparently decreases up to a certain point and then rises quite definitely, so that in the late lesions the proportion of free to ester cholesterol is much greater than in the early lesions (the average ratio 1 in normal, 0.56 in early, 0.73 in medium, and 1.04 in late lesions). This was a surprise and contrary to Schönheimer's finding of a steadily increasing ratio of ester to free cholesterol with progressive pathologic changes. Our figures seem, then, to bear out more fully Aschoff's infiltration theory: that cholesterol esters along with other fats are laid down in the aorta from the blood—merely a physical deposition without any selective process on the part of the aorta. Later the cholesterol esters are split, leaving increasing amounts of free cholesterol.

The phospholipids remain practically constant in the three types of plaques and their average value (17 per cent of the total fatty extract) is much greater than that reported by Schönheimer for his series, but this is undoubtedly due to a difference in extraction. In a recently published paper on the analysis of alcoholic extracts of whole fixed aortas Lehnher<sup>7</sup> reported phospholipid values similar to ours. Normal tissue from young aortas shows a higher phospholipid content than the damaged tissue shows, whereas normal tissue from two old aortas (aged 60) shows phospholipid values similar to those of the plaques and, at the same time, higher total cholesterol values than found for young normal tissue. This is interesting as a suggestion of the effect of age alone on the fat content of the aorta.

Some of the variations in our figures are undoubtedly due to the difficulty of an absolutely clean separation of the intimal tissue involved

7. Lehnher, E.: New England J. Med. 208:1307, 1933.

in lesions from the surrounding tissue, and also to the fact that any individual class of lesions appears to be influenced by the state of the aorta as a whole. For example, in several cases in which both early and late, or both medium and late lesions were taken from the same aorta the percentage of fatty extract and of total cholesterol in the early or medium lesions was above the average for the group, tending to approach the value found for the late lesions. It is interesting, too, that one case of hypertension and one of cholelithiasis showed abnormally high values for fatty extract.

#### SUMMARY

Analysis of arteriosclerotic plaques from forty-five human aortas showed a constant amount of phospholipids, an increase of total fatty extract and total cholesterol with increase in the severity of the lesions, and also an increase, in the late lesions, in the ratio of free to ester cholesterol.

## Notes and News

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**University News, Promotions, Resignations, Appointments, etc.**—William Bulloch has retired from the chair of bacteriology at the London Hospital Medical College. The title of emeritus professor of bacteriology has been conferred on Dr. Bulloch.

Arthur W. Wright has been appointed professor and head of the department of pathology at Albany Medical College to succeed Victor C. Jacobsen who has resigned.

Albert E. Casey, Rockefeller Institute for Medical Research, New York, has been appointed associate professor of pathology at the University of Virginia.

Harold W. Stewart has been made associate in pathology in Jefferson Medical College, Philadelphia.

Gene H. Kistler has been appointed associate professor in the department of pathology and bacteriology in the University of Alabama.

According to *Science* the 1934 James E. Stacy Award, consisting of a medal and a sum of money given by the faculty of medicine of the University of Cincinnati for significant contribution to the theory of focal infection in theory or practice, has been bestowed upon Dr. E. R. LeCount, professor of pathology in Rush Medical College, for "his experimental studies on the isolation of streptococci from sore throats and the experimental induction, through their injection, of acute, healing and scarring types of nephritis, identical with the chronic nephritides observed in man."

A grant of \$1,000 has been made by the Simon Baruch Foundation for Medical Research to the pathologic laboratories of St. John's Hospital, Brooklyn, to carry forward an investigation by Theodore J. Curphey on the effect of extracts of fetal endocrine tissue on cell growth.

Herbert U. Williams, professor of pathology in the school of medicine, University of Buffalo, has retired after more than forty years of teaching. His address is 30 Arlington Place, Buffalo, N. Y.

**Society News.**—The forty-fifth annual meeting of the Association of American Medical Colleges will be held at Nashville, Tenn., Oct. 29, 30 and 31, 1934.

At the thirteenth annual convention of the American Society of Clinical Pathologists, held in Cleveland from June 7 to 11, the following officers were elected: president-elect, F. M. Johns, New Orleans; vice-president, B. S. Kline, Cleveland. The Ward-Burdick Medal was awarded to R. R. Kracke for his work on agranulocytic angina. Ludwig Hektoen and Otto Naegeli were elected honorary members.

The American Association for the Study of Neoplastic Diseases will hold its next meeting in Washington, D. C., on Sept. 6, 7 and 8, 1934, at the Mayflower Hotel.

# Abstracts from Current Literature

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## Experimental Pathology and Pathologic Physiology

GROWTH INHIBITOR IN KIDNEY DESICCATES. F. A. MCJUNKIN and C. D. HARTMAN, Am. J. Path. 9:739, 1933.

It is well known that certain organs elaborate chemical regulators, or hormones, which govern growth and metabolism. It has been demonstrated that several of these inhibit karyokinesis of the cells that produce them. This self-regulatory function of the hormones is not known to depend on their metabolic activity. In the case of the parathyroid gland, the hormone made inactive, so far as calcium metabolism is concerned, still inhibits proliferation of the parathyroid cells. The experiments recorded in this paper describe methods for the extraction from the rat's kidney of an inhibitor of renal tubule mitosis. It is not known that this inhibiting agent exercises any control over growth of the body as a whole, or that it plays any rôle in metabolism. Simple aqueous extracts made from the fresh macerated kidney had little influence on the proliferation of renal epithelium. Simple aqueous extracts of desiccated kidney, however, were inhibitory when sufficiently large doses were injected. A more satisfactory potency was obtained by means of acid-alcohol extraction of either fresh or desiccated kidney. The successful extracts were made with dilute acid and 60 per cent alcohol, and at temperatures of 40 C. or less. The acid-aqueous extracts that were tested were without effect on the renal epithelium. They were prepared at 70 C. or more, and the strength of acid was relatively high. Further attempts are being made to purify and concentrate the extract.

### AUTHORS' DISCUSSION AND SUMMARY.

THE REACTION TO FINE AND MEDIUM-SIZED QUARTZ AND ALUMINUM OXIDE PARTICLES (SILICOTIC CIRRHOSIS OF THE LIVER). L. U. GARDNER and D. E. CUMMINGS, Am. J. Path. 9:751, 1933.

Three series of rabbits were given intravenously 1.3 Gm., respectively, of silica particles 1 to 3 microns in diameter, silica particles 6 to 12 microns in diameter and aluminum oxide particles 1 to 3 microns in diameter. The injections were given in divided doses and required from one to four months for their completion. These particles were segregated in different locations according to their size. The largest ones were caught in the pulmonary capillaries, those of intermediate size in the spleen and hepatic lymph nodes, and the finest ones in the liver. The fine particles of silica were the most active and produced a progressive coarsely nodular cirrhosis of the liver attended by extensive destruction of the parenchyma, followed later by regeneration in certain areas. This cirrhosis was the result of a typical hyaline nodular silicotic fibrosis originating in the portal connective tissues. Coarse particles, 10 to 12 microns in diameter, were much less irritating. They excited a simple foreign body type of reaction which progressed very little in practically three years' time. Fine particles of aluminum oxide the same size as those of the smaller silica fraction and introduced in essentially the same quantity were merely phagocytosed and produced no fibrosis in the stroma of any organ where they were deposited. These observations support the point of view that the injury produced by silica is specific and chemical rather than physical.

### AUTHORS' SUMMARY AND CONCLUSIONS.

PROLONGATION OF PREGNANCY IN THE RABBIT BY INDUCED OVULATION. F. F. SNYDER, Bull. Johns Hopkins Hosp. 54:1, 1934.

The rôle of corpora lutea in parturition was studied by induction of ovulation during pregnancy with urine extract so that at term there was a fresh set of

corpora lutea at the stage of maximal activity. Under these conditions the following observations were made: Normal parturition at term was never observed; in most animals, pregnancy was prolonged, the onset of parturition being delayed until fifteen days after injection (i. e., the fortieth day) or until the end of the life span of the induced corpora lutea. Retention of postmature fetuses longer than the life span of the induced corpora lutea was associated with injury of the uterine wall. Solution of pituitary administered at term failed to induce parturition in a dosage one thousand times greater than the amount normally effective. Fetuses survived in the uterus three days past term, i. e., thirty-five days, and developed to excessive size. An estimate of the functional reserve of the placenta at term was afforded by the extent of the development of postmature fetuses and the persistence of glycogen in the placenta at forty-one days. By varying the stage of pregnancy at injection, the dosage and the parity of the animal, a method was found by which pregnancy was prolonged in twenty of the twenty-three animals, while abortion of the litter occurred in only three. In abortion the fetuses were expelled either alive or before any maceration had taken place. The abortions were limited in their occurrence to the second or the third day after injection. There was no support for the view that the onset of parturition is caused by changes in the fetus, senility of the placenta or mechanical distention of the uterus. Retention of the fetus in the uterus, in the rabbit at least, is under hormonal control. Termination of its retention coincides with termination of the life cycle of the corpus luteum.

#### AUTHOR'S SUMMARY.

EXPERIMENTAL EXOPHTHALMOS AND HYPERTHYROIDISM IN GUINEA-PIGS.  
H. B. FRIEDGOOD, Bull. Johns Hopkins Hosp. 54:48, 1934.

Simultaneous studies of the basal metabolic rate and pathologic changes of the thyroid gland were carried out on thirty guinea-pigs given extracts of the anterior lobe of the pituitary gland over a period extending from forty-eight hours to one hundred and ninety days. Six guinea-pigs were used for controls. The available data suggest the following conclusions:

The injections produce a syndrome which is remarkably similar to that of exophthalmic goiter in man, but there is no evidence that the two conditions are identical in their pathogenesis.

The behavior of the basal metabolic rate during a prolonged period of injections is extremely variable. It is characterized by an initial increase followed by a spontaneous remission, and may exhibit subsequent recrudescences. The spontaneous remission in the basal metabolic rate may be due to a relative functional insufficiency of the thyroid gland (rapid depletion of the stored precursors of the calorogenic hormone with or without parenchymal injury), although the development of a protective "antipituitary" or "antithyroid" substance must be considered a possibility.

Parenchymal hypertrophy and hyperplasia of the thyroid gland appear simultaneously with systemic evidence of its hyperactivity, but these same pathologic changes may still be present when the basal metabolic rate is spontaneously decreasing. In the latter event, this pathologic picture is associated with a decreasing activity of the thyroid gland.

Exophthalmos is produced independently of the calorogenic hormone of the thyroid gland. Anterior pituitary extract seems to be more capable of inducing prominent exophthalmos in the absence of hyperthyroidism and in the presence of hypothyroidism. Chronic exophthalmos which persists after the injections are discontinued may result from prolonged administration of the extract.

#### AUTHOR'S SUMMARY.

THE NERVOUS SYSTEM IN DEFICIENCY DISEASES. H. M. ZIMMERMAN and E. BURACK, J. Exper. Med. 59:21, 1934.

Adult dogs maintained on an artificial balanced ration adequate, so far as is known, in all dietary essentials except water-soluble, heat-stable vitamin B<sub>2</sub> (G)

presented, after a sufficient time, a slowly progressive disease characterized by loss of weight, persistent vomiting and diarrhea, and marked muscular weakness, which ended fatally in from two hundred to over three hundred days. The clinical features of this condition are quite different from those characterizing the canine disease known as black tongue. The anatomic changes consist of marked demyelination of the peripheral nerves, including the vagus; degeneration of the medullary sheaths and replacement by gliosis of the posterior columns of the spinal cord, particularly the fasciculi graciles; degeneration of the medullary sheaths of the posterior and less often of the anterior nerve roots of the cord; occasionally slight degenerative changes in most of the other fiber tracts of the cord. Attention is called to the fact that degenerative lesions in the central nervous system similar to or identical with these have frequently been described in pellagra in man.

#### AUTHORS' SUMMARY AND CONCLUSIONS.

**NUTRITIONAL MYOPATHY IN DUCKLINGS.** A. M. PAPPENHEIMER and M. GOETTSCH, *J. Exper. Med.* **59**:35, 1934.

Ducklings fed on a diet of skimmed milk powder, casein, corn-starch, lard, cod liver oil, yeast, salts and paper pulp rapidly acquire a disease characterized by extreme and progressive myasthenia, ending in death within a few days. Pathologic changes are found in the skeletal muscles. These show widespread hyaline necrosis of fibers, with edema and cellular reaction. The brain and other parts of the central nervous system are not affected, and no significant alterations are found in other viscera or tissues. The creatine content of the muscles is reduced in proportion to their injury. Controls on a natural food diet remain free from the disease.

#### AUTHORS' CONCLUSIONS.

**THE HYPOPHYSIS IN SO-CALLED CONSTITUTIONAL OBESITY.** E. ZEYNEK, Frankfurt. *Ztschr. f. Path.* **44**:387, 1933.

In 81 per cent of thirty-two cases of obesity the hypophysis showed an absolute increase of the basophilic cells. The opinion is stated that the basophilic pituitary cells have a definite relation to the fat metabolism. This view is regarded as partly supported by the fact that in Cushing's syndrome of pituitary basophilism, obesity plays an outstanding part.

WILLIAM SAPHIR.

**FUNCTIONAL DISTURBANCES OF THE PERIPHERAL CIRCULATION.** B. FISCHER-WASELS, Frankfurt. *Ztschr. f. Path.* **45**:1, 1933.

In this article, which comprises the entire first part of this volume, Fischer-Wasels discusses the more important questions and the more modern researches in this field. Ricker's law of the circulation is rejected, and the belief is expressed that it is not a law but merely a rule. It is formulated in that a slight irritation produces, as a rule, an active hyperemia and acceleration of the current. A medium irritation produces, as a rule, a narrowing of the arterioles and a slowing of the circulation. A severe irritation produces, as a rule, a more marked slowing of the circulation, a maximum dilatation of the capillaries and stasis. He points out that the circulation may be impaired because of (1) hindrance of the currents of the blood, (2) damage of the blood corpuscles, (3) changes in the walls of blood vessels and (4) primary tissue changes resulting in local anemia and ischemia or in hyperemia combined with either slowing or increase of the circulation. He believes that spontaneous gangrene may occur without organic occlusion of the vessels. Such a gangrene may be the result of a primary hypersensitivity of the regional circulatory region which produces a standstill of the circulation under the influence of toxic substances or of locally formed metabolic products. The vasomotor nerves do not play a primary principal rôle in causing the gangrene. He stresses how important it is to be critical in the assumption that functional spasms of the vessels explain pathologic observations. Otherwise, much will be explained on the basis of

a hypothesis of arterial spasm. It is emphasized that spasm alone does not cause necrosis. The pathogenesis of brain hemorrhage is also discussed. There is primarily a destruction of brain tissue as a result either of trauma or of toxins (direct effect) or because of local circulatory disturbances (indirect effect). As a result of the primary destruction split products may be present which secondarily damage the walls of blood vessels either by accentuating the local disturbances of circulation or by producing anatomic changes of the vessels which finally may result in necrosis. The damage to the walls of the vessels may also cause reflexory disturbances. The result of the vessel changes may be one massive hemorrhage or multiple simultaneously occurring small hemorrhages. The value of motion pictures of the circulation is stressed.

O. SAPHIR.

### Pathologic Anatomy

**LIPOID HYPERPLASIA OF THE GALLBLADDER.** J. BORENDEN, Arch. f. klin. Chir. **175**:266, 1933.

One hundred and twenty-seven gallbladders that showed signs of lipoid infiltration were studied microscopically. No causative relationship was noted between the storage of the lipoid and gallstones. Nor was there evidence of a primary disturbance in cholesterol metabolism as a basis for lipoid infiltration. There was no connection between the storage of lipoid and inflammation. The accumulation of lipoids results from a deposit of cholesterol esters in the bile. On stagnation of lymph in the mucosa of the gallbladder the cholesterol esters are resorbed and deposited in the tissue.

JACOB KLEIN.

**MULTIPLE CHYLOUS EFFUSION IN BILATERAL PULMONARY FIBROSIS.** P. STEINER, Beitr. z. Klin. d. Tuberk. **81**:757, 1932.

Necropsy on a 32 year old woman who had died with signs and symptoms of cardiac insufficiency revealed chyloous effusions into both pleural cavities, the peritoneum and the pericardium. No obstruction of the thoracic duct could be demonstrated. These findings are explained on the basis of an accompanying extensive pulmonary fibrosis with sclerosis of the pulmonary veins, marked congestion of the lesser circulation, obliteration of the pulmonary lymphatic circulation and fixed diaphragm. Three years previously the patient had had exudative pleurisy on the right side following influenzal bronchopneumonia, and a year later exudative pleurisy on the left side. Both healed by massive fibrosis progressively invading the lungs.

AARON EDWIN MARGULIS.

**TUBERCULOSIS OF THE PERICARDIUM.** ERNST KELLER, Beitr. z. Klin. d. Tuberk. **82**:213, 1933.

Tuberculous pericarditis was found in 130 of 15,659 autopsies in the pathological institute of the University of Heidelberg, an incidence of 0.9 per cent of the total and of 5 per cent of all persons dying of tuberculosis. The disease occurred twice as frequently in males as in females, but without predilection as to age. The associated tuberculosis was miliary and of a predominantly productive form in 12 per cent, and exudative in 88 per cent. The manner of infection of the pericardium was by continuity from lymph nodes and the pleura in 17 per cent, by lymphatic spread in 48 per cent and by hematogenous spread in 35 per cent.

AARON EDWIN MARGULIS.

**ARGENTOPHILIC FIBERS IN THE PRIMARY COMPLEX OF TUBERCULOSIS.** M. D. ARIEL, Beitr. z. Klin. d. Tuberk. **82**:341, 1933.

Ariel studied the histology of the primary complex in sections prepared by impregnation with silver. This method permits greater insight not only into the pathologic anatomy of the lesion but also into the evolution of the process. Forty

primary complexes were so studied, and a comparison of the development of the parenchymal lesion with that of the associated lesion in the lymph node was worked out. It was found that evidences of exacerbations were more frequent in the foci of lymph nodes, with concomitant earlier formation of granulation tissue. By contrast, the parenchymal lesions rarely showed evidence of exacerbations, and the circumfocal granulation tissue more commonly went on to organization and encapsulation.

AARON EDWIN MARGULIS.

LYMPHATIC CYSTS IN A HILAR GLAND. FRIEDRICH STEFFEN, Beitr. z. Klin. d. Tuber. **82**:500, 1933.

Cystic transformation of the hilar and tracheobronchial glands, particularly of those at the bifurcation of the trachea, was found at necropsy in a patient with pneumonoconiosis. The cyst spaces were dilated lymphatic sinuses and lymphatic vessels. The cause was probably the impeded flow of lymph consequent to anthracosis of the lymph nodes. This is said to be the first observation of its kind reported.

AARON EDWIN MARGULIS.

THREE CASES OF MARKED CIRCUMSCRIBED ATROPHY OF THE LIVER. J. JESCHEK, Beitr. z. path. Anat. u. z. allg. Path. **89**:233, 1932.

Atrophy of one lobe of the liver with almost complete destruction of the parenchyma was found at autopsy in three women. In two of the women the finding was accidental and without relation to the cause of death, while in the other Banti's disease was diagnosed before death. The author regards the atrophy as a reparative stage of subacute yellow atrophy of the liver.

C. ALEXANDER HELLWIG.

MORPHOLOGIC EVIDENCE OF THE FUNCTIONAL STATE OF THE SUPRARENAL MEDULLA. M. STAEMMLER, Beitr. z. path. Anat. u. z. allg. Path. **91**:30, 1933.

By morphologic study of the medulla of the suprarenal gland in mice, rats and guinea-pigs after these had been subjected to a variety of experimental procedures that the physiologist has found to cause increased or decreased liberation of epinephrine Staemmler attempted to correlate the morphologic changes in the medulla with its functional state. Granting that the chromaffin substance bears a direct relation to the formation of epinephrine, he finds that the quantity of chromaffin substances does not give a true measure of the functional activity of the medulla. Vacuolation of the cells is of greater importance and indicates increased cellular activity and increased secretion. An increase in the chromaffin content may or may not be associated with vacuolation. The combination of poverty in chromaffin content and vacuolation is interpreted as evidence of hyperfunction and indicates both increased formation and increased liberation of epinephrine. Decrease in chromaffin substance without vacuolation may be due to increased liberation of epinephrine without increased secretion or to a primary disturbance of secretion. Either or both of these physiologic processes sooner or later lead to exhaustion of the medulla. The same physiologic process may yield evidence first of hyperactivity and then of exhaustion. The liberation of epinephrine is under the influence of the splanchnic nervous system, but the formation of epinephrine appears to be less definitely under nervous control. Morphologic evidence of hyperfunction was observed after unilateral extirpation of a suprarenal gland or ligation of its vein, after action of heat, after withdrawal of blood, and in the early stages of morphine, carbon monoxide and phosphorus poisoning. In these intoxications hyperactivity gave way to exhaustion. Peritonitis, diphtheria toxin and application of cold did not lead to increased secretory activity of the medulla. Liberation of the formed store of epinephrine was followed almost immediately by exhaustion.

O. T. SCHULTZ.

**MASSIVE HEMORRHAGE FROM THE VESSELS OF THE NECK IN SCARLET FEVER.**  
T. STEIN, Beitr. z. path. Anat. u. z. allg. Path. 91:202, 1933.

Massive hemorrhage from the vessels of the neck is one of the rarer complications of scarlatina. In 630 cases of scarlet fever which came to necropsy in the Wassiljeostrow hospital for infectious diseases of children in Leningrad this complication was the cause of death 14 times. An additional case from another institution is included. In 10 instances the hemorrhage was arterial; in 8 of these a careful microscopic examination was made. In 5 instances the bleeding was of venous origin; a detailed histologic report of 4 of these is presented. Such hemorrhages are due to destruction of the wall of the vessel by necrosis or acute inflammation, the process reaching the wall from the adjacent diffusely inflamed tissues. In some instances the inflammatory process that encroached on the wall was gangrenous and appeared to be due to secondary invasion by fusiform bacteria; in other instances streptococci appeared to be the cause of the inflammatory process, which was then more purulent. The internal carotid artery was the site of rupture and hemorrhage in 3 cases; in these the phlegmon or abscess was retropharyngeal. The external carotid artery was involved in 1 case and the external maxillary artery in 2; in these the abscess was submaxillary. In 2 cases the common carotid artery, and in 3 the jugular vein, was the source of the hemorrhage, the abscess being situated in the lateral portion of the neck. Hemorrhage was from the anterior jugular vein in 2 instances; the abscess was situated in the anterior portion of the neck. In 2 cases in which the hemorrhage came from an ulcerated, necrotic lesion of the pharynx, the superior laryngeal artery was involved once and the ascending palatine artery once. In half the cases of arterial hemorrhage a single massive bleeding led immediately to death. In the other half the fatal massive bleeding was preceded by one or more smaller hemorrhages. The venous hemorrhages were usually multiple, and death occurred some hours after the terminal massive hemorrhage. In no instance did the fatal hemorrhage occur before the eighteenth or later than the thirty-fourth day of the disease.

O. T. SCHULTZ.

**LAMBL'S EXCRESENCES OF THE AORTIC VALVE.** W. GÜNZEL, Beitr. z. path. Anat. u. z. allg. Path. 91:305, 1933.

The minute excrescences of the ventricular surface of the segments of the aortic valve first described by Lambl in 1858 were found in 50 per cent of ninety consecutive necropsies. Their frequency increased with length of life. In twenty-six of the necropsies the excrescences were subjected to microscopic examination. They were composed of dense, avascular tissue, with no inflammatory cells or other evidence of inflammatory reaction. The collagen and elastic fibrils of the valve could be traced directly into the excrescence. These findings lead Günzel to deny the correctness of the two usually accepted views of the origin of the outgrowths: that they are organized small thrombi or that they are the result of subendocardial inflammation of the valve. He believes that their origin is purely mechanical and that they result from rupture of a few collagenic and elastic fibrils caused by the changes in pressure that occur with systole and diastole. The ruptured fibrils buckle outward and are then elongated into the typical excrescences by the continuing action of the blood stream.

O. T. SCHULTZ.

**THE RETICULUM OF THE SPLEEN IN CIRRHOSIS OF THE LIVER.** L. JORES, Beitr. z. path. Anat. u. z. allg. Path. 91:343, 1933.

The reticulum of the spleen was studied by a modification of Leo Müller's method of preliminary digestion of paraffin sections by pancreatin. In central congestion of the liver of cardiac origin no increase of reticulum was detected,

but portions of individual fibers were thickened. In cirrhosis of the liver two types of alteration of the splenic reticulum were noted. In one there was periarterial and follicular fibrosis with hyperplasia of the sinusal reticulum. In the other the hyperplasia was limited to the pulp. The first type was associated with interlobular cirrhosis without jaundice and without hematogenous pigment in the liver; the second, with cirrhosis in which there were intralobular fibrosis and hematogenous pigmentation. The reticular hyperplasia of the spleen was often focal and unsymmetrical. The changes in the reticulum are ascribed to alterations in the function of the spleen.

O. T. SCHULTZ.

**CHANGES IN THE KIDNEYS IN GOUT.** T. FAHR, Centralbl. f. allg. Path. u. path. Anat. **57:49**, 1933.

Fahr reports the changes in the kidneys of a man, 74 years old, who had gout for thirty years and died of sclerosis of the coronary artery and heart failure. The kidneys weighed 60 Gm. each and were granular and gray-brown; the fat of the hilus was increased. The cortex was narrow. There were huge quantities of urates in the interstitial tissue of the medulla, and some smaller collections were separated by bridges of connective tissue. In seeking an explanation for this sharp localization of urates, Fahr considers first a hematogenous source. To reconcile this mode of transport with the localized deposition he believes that some special predisposition of the medulla is necessary, similar to that in the ear. As a second possibility, he considers that the urates may be carried in the lymphatics from the degenerated collecting tubules.

GEORGE RUKSTINAT.

**THE EPITHELIUM OF THE PULMONARY ALVEOLI.** F. ORSÓS, Centralbl. f. allg. Path. u. path. Anat. **57:81**, 1933.

Orsós believes that there is a continuous respiratory epithelium lining the alveoli. This is best studied in sections from 3 to 5 microns thick, allowing a view of the individual cells, or in sections from 50 to 200 microns thick, which convey the idea of a granular, fenestrated membrane. The cells comprising this structure are of varying types. One type is polygonal or rounded and has a large pale nucleus and radial striations in the periphery of the granular cytoplasm. Another has nuclear protoplasm, and another pseudopodium-like processes. Inclusions, such as coal pigment, drops of fat, masses of fibrin or red blood cells, frequently occur in the membrane. The membrane may be loosened during life by inflammation, as by an exudate under it. Postmortem hypostasis may similarly effect a separation. When the membrane is detached, it may imprison some air and in shrinking may resemble a punctured balloon. Further shrinking may result in a fine veil-like cellular mass. When the individual cells die they go over into the anuclear part of the membrane.

GEORGE RUKSTINAT.

**MECKEL'S DIVERTICULUM AND GALLSTONES.** H. HANKE, Centralbl. f. allg. Path. u. path. Anat. **57:161**, 1933.

Fifteen faceted gallstones, each about half as large as a pea, were found in a Meckel diverticulum in a man who had died of heart failure. The stalk of the diverticulum had a large lumen, and its lining had a fibrinopurulent coating. The gallbladder contained many stones, one the size of a hen's egg, and was chronically inflamed. The passage of stones from the gallbladder to the diverticulum was facilitated by an enlargement of the ductus choledochus and of the orifice of the diverticulum.

GEORGE RUKSTINAT.

HEMORRHAGIC INFARCTION OF BOTH SUPRARENAL GLANDS IN SEPSIS. S. SCHEIDECKER, Centralbl. f. allg. Path. u. path. Anat. 57:163, 1933.

A boy, 6½ years old, fell, striking his head on a stone. Meningitis developed within two days, and the boy died shortly after. The periosteum at the site of injury was intact, and there were slight evidences of meningitis. Both suprarenal glands contained free blood, especially in the medulla, and the capillaries contained emboli in which were diplococci and streptococci. Similar organisms were found in the cerebral exudate. Death was due to acute suprarenal insufficiency.

GEORGE RUKSTINAT.

AN UNUSUAL CAUSE OF SUDDEN DEATH IN SYPHILITIC AORTITIS. H. SIKL, Centralbl. f. allg. Path. u. path. Anat. 57:228, 1933.

A tailor, 54 years old, died suddenly after complaining for a few hours of pain in the chest. At necropsy a fresh, clot-covered gumma was found at the mouth of the left coronary artery. Microscopically there were caseous necrosis of the media and thickening of the intima near the lesion. Spirochetes were found in the zone of acute inflammation, and there leukocytes were abundant.

GEORGE RUKSTINAT.

SPONTANEOUS RUPTURE OF THE GALLBLADDER IN SCARLET FEVER. H. SAWRIMOWITSCH, Centralbl. f. allg. Path. u. path. Anat. 58:49, 1933.

A tear 4 cm. long was found in the gallbladder of a boy, 6 years old, who died on the eighteenth day of scarlet fever. The gallbladder measured 12 by 4 cm., and the tear was on the left side of the fundus. There was no peritoneal reaction to the sterile bile, although rupture of the bladder had presumably occurred some days before death. At the edges of the rent were partially organized thrombi and hyperplastic epithelium. It is supposed that the rupture occurred in a markedly distended organ, because the latter was large, was not inflamed and was ruptured at the fundus. The overdistention is explained on the basis of paresis of the wall of the bladder due to toxemia. This, coupled with a slight mechanical obstruction to the outflow of bile, such as enlarged lymphatic glands about the duct or some dyskinetic alteration of the sphincter, might lead to rupture.

GEORGE RUKSTINAT.

A PERFORATING ANEURYSM OF THE LEFT VERTEBRAL ARTERY CAUSED BY GUMMATOUS DISEASE OF THE VASCULAR WALL. A. ESSER, Frankfurt. Ztschr. f. Path. 43:448, 1932.

Autopsy of a woman whose condition had been diagnosed clinically as dementia paralytica revealed atrophy of the brain, a perforated aneurysm of the left vertebral artery, syphilitic aortitis and moderate sclerosis of the aorta. Histologic examination disclosed syphilitic meningo-encephalitis and gummas in the left vertebral artery. Esser discusses the etiology of the aneurysm and the differential diagnosis of syphilitic endarteritis and gummas of the walls of the blood vessels. Because the lesions were found in the intima and because giant cells were not present in them, it was at first difficult to decide whether they were gummas or whether they could be explained on the basis of a syphilitic endarteritis.

O. SAPHIR.

CONGENITAL AORTIC STENOSIS COMBINED WITH ENDOCARDIAL HYPERPLASIA IN THE NEW-BORN. E. DISSMAN, Frankfurt. Ztschr. f. Path. 43:476, 1932.

A case is reported. The aortic valve in the region of the bulbus revealed three areas of thickening parallel to the aorta. One of the areas formed a ball-like structure measuring about 4 mm. in diameter, which almost completely obstructed the lumen of the aorta. The endocardium of the left ventricle correspond-

ing to the interventricular septum just beneath the aortic valve was white and measured as much as 2 mm. in thickness. Its surface was smooth in some portions and rough in others, resembling the trabeculation of the apex of the left ventricle. Dissman believes that the thickenings of the endocardium and in the region of the aortic valve were the result of primary hyperplasia of the embryonal texture of the valvular apparatus and of the endocardium. He does not believe that an intra-uterine inflammation was responsible for these changes.

O. SAPHIR.

HISTOLOGY OF THE INFLAMMATORY EXUDATE IN GRANULOCYTOPENIA ("AGRANULOCYTOSIS"). K. VAN DER WIELEN, Frankfurt. *Ztschr. f. Path.* **44**:34, 1932.

Eleven cases of granulocytopenia are described. The foci of inflammation revealed that the inflammatory cells corresponded to the cells of which the blood consisted in so far that in instances in which no granulocytes were present in the circulating blood polymorphonuclear leukocytes could not be demonstrated in the areas of inflammation, while in instances in which polymorphonuclear leukocytes were markedly reduced in number in the circulating blood, only a few were found in such areas. Many macrophages and a marked histiocytic reaction, however, could be demonstrated. Since the changes in the bone marrow were not uniform, so-called agranulocytosis does not seem to be a primary well characterized disease of the blood-forming marrow. There seems to be no connection between granulocytopenia and syphilis. Occasionally, however, antisyphilitic treatment may be of value. It is concluded that the dependence of the inflammatory exudate on the types of cells in the blood supports Fischer-Wasels' theory, according to which the leukocyte seen in areas of inflammation is an emigrated blood cell and not the product of local cell metamorphosis.

O. SAPHIR.

INFLUENCE OF ROENTGEN RAYS ON TRAUMATIC INFLAMMATION. H. BUHTZ, Frankfurt. *Ztschr. f. Path.* **44**:57, 1932.

Skin wounds treated with roentgen rays revealed twenty-four hours later a greater tendency toward healing than control wounds. One hour after roentgen treatment the migration of leukocytes in the region of the wounds was much more marked in the treated animals than in the nontreated controls. Four hours after treatment the maximum of the leukocyte migration was reached in the treated wounds. Twenty-four hours after treatment the accumulation of leukocytes was confined to the most superficial portions of the treated wounds, while hardly any leukocytes could be made out in the tissue surrounding these wounds. In the control wounds the peripheries showed a marked infiltration of leukocytes, which were also present a short distance away from the wounds. A transformation from connective tissue cells into leukocytes could not be demonstrated.

O. SAPHIR.

### Pathologic Chemistry and Physics

STUDIES IN OSCILLOMETRY. ALFRED FRIEDLANDER, Am. Heart J. **9**:212, 1933.

The form of the oscillogram, taking curves of all extremities in each case, is of more importance than the estimation of the maximal oscillometric phase. The normal oscillogram may show definite variations in the maximal oscillometric phase (MOP) in the same patient at different times. This is dependent largely on the condition of peripheral resistance. Typical pathologic oscillograms are shown, demonstrating arterial thickening, essential hypertension with nephrosclerosis, and hypertensive heart disease with Mönckeberg's sclerosis. From the form of the oscillogram in typical cases definite conclusions may be drawn as to the condition of the vascular tree. Mixed forms of curve are often found. In such cases, the oscillogram alone cannot afford a definitive diagnosis of the condition of the walls of vessels.

AUTHOR'S SUMMARY.

**THE CREATINE CONTENT OF THE MYOCARDIUM OF NORMAL AND ABNORMAL HUMAN HEARTS.** DONALD W. COWAN, Am. Heart J. 9:378, 1934.

The average creatine content of the left ventricular myocardium as determined in forty-eight approximately normal human hearts was 194 mg. per hundred grams of tissue, this value being based on an arbitrary water content of 80 per cent. There were no differences in this creatine content related to either sex or age. A few hearts included in the normal group, but having creatine contents differing widely from the mean, are listed separately and the diagnoses given in detail. Analyses of seventeen decompensated hearts showed an average creatine content significantly lower than normal. Scar tissue was not a significant factor in producing this low value. Fifteen abnormal, but not decompensated, hearts had an average creatine content significantly lower than normal, but higher than the values for the decompensated hearts. These are listed separately, with their diagnoses given in detail. Septicemia *per se* had no effect on the creatine content of the left ventricle. An effect of hypertrophy *per se* on the creatine content of the heart was not definitely established. These findings suggest that the "reserve" of the heart closely parallels its creatine content.

**AUTHOR'S SUMMARY.****BLOOD LIPIDS IN CHILDREN WITH SCARLET FEVER AND RHEUMATIC DISEASE.** A. D. KAISER and M. S. GRAY, Am. J. Dis. Child. 47:9, 1934.

Studies on the blood lipids of children with rheumatic disease and with scarlet fever revealed values similar to those found in normal children. The standard deviation from the mean was considerably greater in the children with these infections than in normal children. It is likely that the infection rather than the varied amount of fat ingested was responsible for the increased deviations.

**STEATORRHEA IN PANCREATIC DISEASE AND SPRUE.** JOSEPH H. PRATT, Am. J. M. Sc. 187:222, 1934.

The fat content of dried feces was determined in thirty-three cases. In twenty-five cases the fat present constituted 30 per cent or more. In fifteen cases, 50 per cent or more of the dried feces was composed of fat. In these instances steatorrhea was due to obstructive jaundice, obstruction of the pancreatic ducts or sprue, except in three cases in which the cause of the fatty diarrhea was not discovered. The absorption of fat was studied in sixteen cases, the standard intestinal test diet of Adolf Schmidt being used. The fat lost in the feces ranged from 2.1 per cent in a patient with normal digestion to 66.2 per cent in a man with obstruction of the common bile duct and the pancreatic ducts. In the cases of steatorrhea, with two exceptions, 24 per cent or more of the fat of the food was excreted in the feces. The absorption of the nitrogen of the test diet was determined in this group of sixteen cases. In obstruction of the pancreatic ducts there was a loss of nitrogen in the stools, indicating faulty digestion and absorption of protein. When fat loss was due to absence of bile from the intestine the absorption of nitrogen remained normal. In sprue, the absorption of nitrogen is usually normal, but may be disturbed. The utilization of the starch in the food was normal in all the cases of steatorrhea studied. The chemical analysis of the feces for fat is important in determining, even approximately, the fat content of the stools, as the results of the microscopic examination of the fresh feces may be misleading. The determination of the percentage of nitrogen in the feces has proved of no value in diagnosis. To study adequately cases of fatty diarrhea it is necessary to determine the absorption of both nitrogen and fat with the patient on a weighed diet, preferably the standard intestinal test diet of Schmidt. Excluding the cases of steatorrhea due to absence of bile from the intestine, most of the instances studied in the present series have proved to be cases of sprue or pancreatic disease. In this study three cases of steatorrhea of unknown origin have been encountered. Further work is needed on similar cases. This should include, in addition to the

clinical study, a complete quantitative chemical analysis of the feces for the remains of food elements and the determination of the amounts of fat and nitrogen of the food that are lost in the stools, and finally a thorough study of the abdominal organs at autopsy.

## AUTHOR'S SUMMARY.

COPPER AND IRON IN CELLULAR METABOLISM. A. LOCKE, D. O. ROBASH and L. E. SHINN, *J. Infect. Dis.* **54**:51, 1934.

A method is described for the approximation of that portion of the total amount of copper and iron present in tissue which may be presumed to be directly active in the motivation of cellular metabolism. Values are given for the copper and motivating iron contents of the tissues of the rapidly growing normal male rabbit which indicate the presence in the liver, heart, kidney and muscle of a metabolic catalyst containing two parts of reducing iron to each part of oxidizing copper. Approximately ten times as much of the catalyst may be present in the maximally active musculature of the heart as is present in the minimally used, least pigmented muscles of the thighs. The catalyst of the brain, orienting a metabolism more largely devoted to the support of respiration, may contain its active copper and motivating iron in a 1:1 ratio. Prolongation or intensification of the endogenous metabolism in the rabbit as the result of fasting, the feeding of thyroid, or fever is accompanied by a wasting of the tissues and a strain on the catalytic balances. Motivating iron is lost through oxidation, and the copper-ferrous iron ratio of the heart muscle may rise to values approaching those observed in brain. An identical effect is reached, with enormously greater rapidity, following injections of diphtheria toxin and following inoculation of *Pneumococcus* type I. The effect is not obtained in protected animals. The lowered metabolism and increased weakness and susceptibility to infection of rabbits maintained on cabbage are accompanied by a lowering of the copper levels. No specific changes in the copper and motivating iron contents of the tissues of the chicken accompany the growth of the Rous sarcoma. Changes are observed during the degeneration of the tumor. The metabolic catalyst of the tumor itself appears to contain less active copper per unit of active iron content than is present in the invaded tissue.

## FROM AUTHORS' SUMMARY.

FURTHER OBSERVATIONS ON THE ELECTRIC CHARGE OF THE ERYTHROCYTES IN CERTAIN PROTOZOAL DISEASES. H. C. BROWN, *Brit. J. Exper. Path.* **14**:413, 1933.

It has been shown that the electric charge of the erythrocytes in malarial birds bears a definite relation to the degree of infection, and that the reduction in charge of the red cells of an infected bird is apparently an important factor in the extent to which phagocytosis takes place. In other words, when the bird with malaria shows a strong immunizing response in that it is successfully combating its infection, the negative charge of the red cells is considerably lowered, but in the bird in which the parasites continue to increase steadily the charge does not markedly differ from that of the red cells in a normal bird. This reduction in the negative charge of the red cells also takes place, but to a less extent, in the blood of mice recovering from *Trypanosoma equiperdum* infection, the change occurring at the time when antibodies appear in the peripheral blood. It has been shown that the reduction in the negative charge of the red cells is nonspecific, and due to the action of the euglobulin fraction of the serum on the erythrocytes concerned. It is well known that in phagocytosis of bacteria these enter the phagocytes when they have their negative charge reduced in the presence of a suitable electrolyte and immune serum. When one takes into account the protein changes in malaria, with the charge-reducing action of euglobulin, it seems conceivable that the rationale of the treatment of dementia paralytica by malaria might be the phagocytosis of the spirochetes induced by an increased euglobulin content of the patient's serum.

Caldwell, in discussing this subject ("Treatment of General Paralysis by Malaria"), stated that the improvement which takes place is primarily due to a destruction of the spirochetes, and has shown that not only the serum but also the cerebrospinal fluid of paralytic patients who have been treated with malaria shows a higher content of agglutinin for the spirochete than is shown by the serum or spinal fluid of untreated patients. If, then, the agglutination of the spirochete occurs during malaria therapy, it follows that the charge of the organism is lowered, and conditions are therefore more favorable for phagocytosis.

Although a considerable amount of work has been done on the sedimentation rate of the red blood corpuscles in various morbid conditions, no very definite explanation appears to have been given to account for the variations that occur. Jones concluded that either an increase of fibrinogen or a decrease in the ratio of albumin to globulin is responsible for an increased rate. It appears that in such diseases as kala-azar, malaria, syphilis and trypanosomiasis the electric charge of the red cell, and consequently the sedimentation rate, is dependent on the proportion of the various proteins present in the plasma, and that such changes in the protein composition of the cerebrospinal fluid are responsible for the lowering of the charge and consequent phagocytosis of the spirochetes in cases of dementia paralytica undergoing malaria therapy.

FROM AUTHOR'S DISCUSSION.

### Microbiology and Parasitology

TYPHUS IN MICE. J. LAIGRET and J. JADIN, Arch. Inst. Pasteur de Tunis **21**:381, 1933.

Mexican virus transferred through mice by injecting brain containing it into their peritoneal cavities was successfully maintained for sixteen passages. Then the series was interrupted. Brain to brain passages were less successful, the virus failing after the fourth passage. With the human virus (Tunis) only several transfers were accomplished by either method of injection. Only silent infections appeared in all cases; the infections were made apparent by guinea-pig inoculations, but even in the guinea-pig controls latent infection was frequent. The survival of Mexican virus in mouse brains reached forty days with intraperitoneal injections, but only ten days with intracerebral injections.

M. S. MARSHALL.

STRUCTURE AND NATURE OF THE SO-CALLED PROTOZOAL BODIES IN THE EXCRETORY DUCTS OF THE SALIVARY GLANDS. G. C. PARENTI, Sperimentale, Arch. di biol. **11**:157, 1933.

The salivary glands of 200 persons, mostly children and fetuses, were examined microscopically. It was possible to demonstrate in 15 cases the protozoa-like cells in the excretory ducts. None were found in the salivary glands of adults. Parenti doubts that these are parasitic bodies, but believes rather that they are acinar cells which have assumed an atypical position and growth.

JACOB KLEIN.

BIOLOGY OF THE TUBERCLE BACILLUS. BRUNO LANGE, Beitr. z. Klin. d. Tuberk. **81**:235, 1932.

Lange reviews the newer work on the biology of the tubercle bacillus. He concludes that the greatest advances have been made in the technic of its isolation and culture and in appreciation of its virulence. He feels that little of the evidence relating to the supposed variation in form, filtrability and dissociation of the classic type is convincing. Restudy of the question of the pathogenicity of the bovine strain for man has reaffirmed that potentiality. He leans to the view that the most frequent way in which man in general is infected is by inhalation of few bacilli.

AARON EDWIN MARGULIS.

## Immunology

SO-CALLED "THYMIC DEATH." G. L. WALBOTT, Am. J. Dis. Child. **47**:41, 1934.

Of a group of 102 cases in which the diagnosis of status thymicolymphaticus was made by various pathologists, 34 were selected in which no adequate cause of death had been determined. The remaining 68 cases were excluded, since other conditions, such as hyperthyroidism, trauma at birth, gastro-enteritis and respiratory infections, were associated with the lymphoid hyperplasia and in themselves were sufficient to cause death. In 11 of the 34 cases death occurred without previous illness, nor were there any pathologic lesions to indicate the presence of a previous illness. In 16 cases, minor, usually nonfatal incidents preceded death. In seven, a syndrome developed manifested as dyspnea, stridor, fever and shock. The lungs of all these patients presented uniform changes, characterized by capillary congestion, extravasation of blood cells and edematous fluid. These lesions alternated with areas of emphysema and atelectasis. In 17 cases petechial hemorrhages were present, involving the heart, pleura and lymph glands and various other viscera. In some, there were dilatation of the right side of the heart and degenerative changes in the liver and edema and capillary congestion in other organs than the lungs. Hypoplasia of the suprarenal glands and hyperplasia of the lymphoid organs, as noted by other authors, were present. Comparison of this pathologic process with that reported in anaphylactic shock in man reveals a close resemblance, if not a complete identity, of the two conditions. Eosinophilia of the tissues and an allergic familial or personal history point further to this conception. On the basis of these findings it is believed that death may be the result of a primary anaphylactic edema of the lungs and ensuing asphyxiation. Such a theory can be upheld only if one assumes (1) that anaphylactic shock may occur from incorporation into the body of nonprotein substances, and (2) that absorption of shock-producing antigen may take place by ways other than by injections. Evidence is presented both from personal experience and from that of others which tends to confirm these facts.

### AUTHOR'S CONCLUSIONS.

PROTEINS AND AN ALCOHOL-PRECIPITABLE CARBOHYDRATE FRACTION OF GONO-COCCI AND MENINGOCOCCUS. A. K. BOOR and C. P. MILLER, J. Exper. Med. **59**:63 and 75, 1934.

No essential differences between the nucleoproteins and the intact cells of Gonococcus and Meningococcus were observed in their ability to engender immune substances (precipitins), to induce bacterial allergy in rabbits or to elicit cutaneous reactions (of the delayed type) in rabbits rendered hypersensitive to these organisms. Measured by their lethal action in mice, the toxicity of gonococcal and meningococcal nucleoproteins was found to be but slightly less than that of the intact cells. It seems probable, therefore, that the toxic action of these organisms is due chiefly or entirely to some constituent of the nucleoprotein. Extraction with acetone and ether in the cold did not reduce appreciably the toxicity of these organisms and their nucleoproteins, nor alter their immunologic behavior. Cross-precipitin reactions suggested that gonococcal nucleoprotein has an antigenic factor in common with the nonencapsulated pneumococcus cell, and meningococcal nucleoprotein, one in common with the capsular material of Pneumococcus type III. Tryptic digestion destroys these antigenic factors, but not those responsible for the cross reactions within the genus Gonococcus.

The alcohol-insoluble polysaccharides of Gonococcus and Meningococcus were found to contain 4.2 and 3.7 per cent nitrogen respectively, to be protein-free by chemical test, and to reduce Fehling-Benedict's solution only after hydrolysis. They were nontoxic for rabbits and mice and failed to engender antibodies (precipitins) in rabbits. They produced no cutaneous reactions in normal, snuffles-free rabbits, but caused typical allergic reactions of the delayed type in rabbits rendered hypersensitive to these organisms. Both carbohydrates reacted in high dilution with

antipseudomococcus serum type III. For comparison, carbohydrates were prepared also from Micrococcus catarrhalis, Streptococcus haemolyticus, Staphylococcus aureus and a rough strain of Pneumococcus.

AUTHORS' SUMMARIES.

THE STUDY OF ANAPHYLACTIC PHENOMENA WITH BLOOD VESSEL PREPARATIONS.

J. WEISSFEILER, *Ztschr. f. Immunitätsforsch. u. exper. Therap.* **79**:452, 1933.

Friedberger advocated the humoral nature of the anaphylactic reactions. Kritschewsky and others who propounded their cellular nature showed that thoroughly washed blood vessel preparations from sensitized animals continued to react to added antigen. That experiment of Kritschewsky was explained by Friedberger as being due to edematous fluid in the wall of the blood vessel and therefore not contradicting the humoral hypothesis. When the preparation is left hanging for from twelve to twenty-four hours, during which time the edema disappears, no reaction occurs when an antigen is introduced. Kritschewsky countered with an experiment which favors the cellular hypothesis: The failure of the blood vessel preparation to react after a long period is due not to the disappearance of the edematous fluid, but to necrobiotic changes. Weissfeiler repeated and confirmed the experiment of Kritschewsky and his associates. A comparison of the reactions of blood vessel preparations of guinea-pigs with those of rabbit's ear showed qualitative as well as quantitative differences. The preparations from the rabbit reacted irregularly and could be desensitized only after repeated additions of the antigen. According to Weissfeiler, the anaphylactic reaction results from a specific rise of an already normally present nonspecific sensitiveness toward the antigen.

I. DAVIDSOHN.

THE CURABILITY OF LATE SYPHILIS IN RABBITS AND THE QUESTION OF IMMUNITY. P. MANTEUFEL and K. HERZBERG, *Ztschr. f. Immunitätsforsch. u. exper. Therap.* **79**:482, 1933.

Two groups of twenty rabbits were used for the experiment; one group had had a syphilitic infection, and the other, an infection with framibesia tropica. A reinfection with the homologous parasites was asymptomatic. Several months after the reinfection the animals were subjected to a thorough treatment with neoarsphenamine. The effect of the treatment was checked by means of glandular transplants which were negative in the cases tested. The rabbits were then reinfected for the third (or in some rabbits for the second) time. The infection was again asymptomatic. Five months later the rabbits were killed and their organs transplanted into the testicles of healthy rabbits. Only twenty-one rabbits survived the entire experiment, and the organs of thirteen of these were free from spirochetes, while the remaining eight still showed the presence of the disease. The thirteen rabbits were considered as immune by Manteufel and Herzberg. That interpretation is contrary to both hypotheses of Kolle, who maintains: (1) that late syphilis in rabbits cannot be cured with arsphenamine, and (2) that in syphilis of rabbits there develops only a "mono-immunity" against the homologous strain as contrasted with the panimmunity which develops in human syphilis. Cross-experiments failed to furnish a means of biologic differentiation between Spirochaeta pallida and Spirochaeta pertenuis.

I. DAVIDSOHN.

IMMUNITY IN SYPHILIS OF RABBITS. H. GROSSMANN, *Ztschr. f. Immunitätsforsch. u. exper. Therap.* **79**:495, 1933.

Twenty-seven rabbits with late syphilis received a course of three injections of neoarsphenamine (0.15 Gm. per kilogram of body weight) and were reinfected after twenty-nine days. Only one animal responded with a local reaction; in the others the infection was asymptomatic. The majority of the animals with asymptomatic infection (eighteen of twenty-three) were free from spirochetes, as shown by transplantation of their organs into healthy rabbits. From the result Grossmann

concluded an immunity existed in late syphilis of rabbits following the treatment with neoarsphenamine. The immunity was of short duration (less than five and one-half months). During the period of absolute immunity the skin of the rabbits was refractory to reinfection with *Spirochaeta pallida*. That observation invalidates, according to Grossmann, the prevalent opinion that cutaneous resistance (anergy) indicates the presence of spirochetes in such rabbits or, in other words, the incurability of late syphilis in these animals.

I. DAVIDSOHN

**RELATION BETWEEN FOOD AND IMMUNE HEMOLYSIS IN RATS.** ARNE FORSSBERG, *Ztschr. f. Immunitätsforsch. u. exper. Therap.* **80**:16, 1933.

Lack of vitamin A had a slight depressing effect on the production of antisheep hemolysins in rats. No such influence was noticed when the other vitamins were removed. An increase of fat in the food had a depressing effect on the production of hemolysins. A similar effect followed the replacement of casein by gelatin and a decrease of protein in the diet. The cholesterol in the blood was inversely proportionate to the antibody titers. The antibody titer rose in response to feeding with lecithin and still more to feeding with choline, which may be the responsible fraction in the lecithin.

I. DAVIDSOHN.

**A CRITICISM OF THE PHAGOCYTIC DOCTRINE.** I. L. KRITSCHEWSKI and W. W. AWRECH, *Ztschr. f. Immunitätsforsch. u. exper. Therap.* **80**:28, 1933.

Metchnikoff postulated a greater potency of phagocytes in immune animals, but he never proved it experimentally. Many investigators after Metchnikoff tried to furnish the experimental evidence and thought that they had succeeded. Kritschewski and Awrech analyze their records and disagree with the conclusions. Their own experiments on phagocytosis in mice immunized with *Spirillum Duttoni* failed to show any evidence of an increase of phagocytic activity in vitro and in vivo as compared with nontreated animals.

I. DAVIDSOHN.

**THE PHENOMENON OF PLASMA COAGULATION IN STAPHYLOCOCCIC INFECTIONS.**

MARIA SUDHUES and R. SCHIMRIGK, *Ztschr. f. Immunitätsforsch. u. exper. Therap.* **80**:42, 1933.

The ability of staphylococci to coagulate human plasma was not influenced by the presence of acute or chronic staphylococcic infections. Similar results were obtained with rabbits infected with virulent staphylococci. The differences in the coagulation of plasma due to the action of staphylococci are not due to antibodies directed against the staphylococci but are probably caused by chemical properties of the plasma. The rapidity of the coagulation of the plasma was inhibited by the addition of bile salts and of lecithin.

I. DAVIDSOHN.

**COAGULATION OF FIBRINOGEN AND "PSEUDO-AGGLUTINATION."** IVAN GYOERFFY, *Ztschr. f. Immunitätsforsch. u. exper. Therap.* **80**:52, 1933.

Bordet and Streng described the phenomenon of conglutination of red blood cells by active ox serum. They attributed it to a special substance in the ox serum which they named "conglutinin." Streng described the same phenomenon in mixtures of certain bacterial species with active ox serum. In a previous report Gyoerffy proved that the phenomenon of conglutination of red blood cells can be satisfactorily explained without the aid of a new immunologic substance (conglutinin): The active ox serum hemolyzes the red blood cells, and during hemolysis, a ferment (kinase) is liberated which brings about flocculation of the residual fibrinogen in the ox serum. In the present report, Gyoerffy shows the validity of the same explanation for the phenomenon of flocculation in mixtures of bacterial suspensions and active ox serum. Flocculation took place only when the ox serum had normal lysine for the bacteria present in the mixture. Extracts

of bacterial bodies were flocculated. Serums of dogs, guinea-pigs and cats were not suited for the experiment. According to Gyoerffy, the results disprove the conglutination hypothesis of Bordet and Streng.

I. DAVIDSOHN.

## Tumors

SARCOMATOID METASTASES IN THE LYMPH NODES DRAINING A PRIMARY CARCINOMA WITH A SARCOMATOID STROMA. R. B. GREENBLATT, Am. J. Path. 9:225, 1923.

A case of sarcomatoid metastases in lymph nodes draining a primary carcinoma with a sarcomatoid stroma is reported. The usefulness of Masson's trichrome blue stain, particularly in the histologic study of the fibroglia of fibroblasts, is emphasized. The possibility of epithelial metaplasia is discussed. An analogy between this case and the experimental tumors of Ehrlich, Haaland and others is assumed. The dividing line between mixed tumors and tumors the stroma of which is not clearly neoplastic is difficult to determine. In this case further metastases by way of the blood stream alone will decide. The occurrence of sarcoma in the lymph nodes may be explained on the basis of the absorption of products from the degenerated epithelium by the lymphatics. This initiates a reaction of the stroma in the lymph nodes which may become neoplastic, depending on the tissue response of the host and the irritative quality of the products liberated by the degenerating cancer cells.

AUTHOR'S SUMMARY.

NEUROBLASTOMA METASTASES IN BONES, WITH A CRITICISM OF EWING'S ENDO-  
THELIOMA. H. C. COLVILLE and R. A. WILLIS, Am. J. Path. 9:421, 1933.

A tumor presenting all the accepted characteristics of Ewing's sarcoma of bone was shown at autopsy to be one of many metastases from a suprarenal neuroblastoma. A review of certain adequately recorded cases of supposed multiple bone sarcoma that came to autopsy leads to the conclusion that these also were instances of suprarenal neuroblastoma with skeletal metastases. The term "Ewing's sarcoma," while possessing clinical value as defining a syndrome presented by a certain group of tumors affecting bones, has no established claim as designating a pathologic entity. It is not denied that there may be a primary bone tumor presenting the Ewing syndrome, but it is believed that further study will disclose the metastatic nature of most of the tumors with this syndrome, and it is strongly suspected that in many of the cases the primary growth will prove to be a suprarenal neuroblastoma.

AUTHORS' SUMMARY.

CALCIFIED EPITHELIOMA OF THE SKIN. K. CH'IN, Am. J. Path. 9:497, 1933.

Ten cases of calcified epithelioma of the skin examined in the pathologic laboratory of the Peiping Union Medical College are reported. Tumors of this type form a distinct group of neoplasms that are anatomically and clinically well defined. They are circumscribed, well encapsulated growths beneath the skin, consisting of lobulated epithelial masses with a network of usually hyalinized fibrous stroma. The epithelial cells are small, oval, deep-staining and closely packed, and have a marked tendency to undergo necrosis, calcification and ossification. A study of the 10 cases and the 116 instances collected from the literature indicates that the growths are distributed most frequently on the head and neck and occur usually in younger persons. The large majority of the tumors are benign, but a few cases of recurrence following removal have been recorded.

AUTHOR'S SUMMARY.

**ANAPHYLACTIC MANIFESTATIONS IN CANCER.** M. CUTLER and W. SAPHIR, J. Allergy 4:389, 1933.

An investigation was made to determine whether cancer produces anaphylactic phenomena. A study of this kind seemed warranted in view of the old clinical observation that many patients with cancer suffer frequently from the so-called anaphylactic diseases, such as hay fever, bronchial asthma, urticaria, angioneurotic edema and vasomotor cutaneous disturbances. Furthermore, there was reason to expect the presence of an anaphylactic condition in the human being with carcinoma since similar conditions have been reported in animals harboring malignant tumors. Two methods of approach were used. First, a search was made for local anaphylaxis. Skin tests with a cancer extract were made on sixty patients. Positive reactions were noted in 62 per cent of the patients with cancer as compared with 24 per cent of the controls. Second, an attempt was made to demonstrate anaphylactic antibodies in the serum of the patients with cancer who showed positive reactions to skin tests. Guinea-pigs previously sensitized with patients' serum were given injections of the cancer extract. These tests gave negative results.

W. SAPHIR.

**MULTIPLE PRIMARY MALIGNANT NEOPLASMS.** H. H. HURT and A. C. BRODERS, J. Lab. & Clin. Med. 18:765, 1933.

Persons with malignant conditions are likely to have more primary malignant neoplasms than a review of the literature would lead one to believe. Multiple primary malignant neoplasm occurs most commonly in the same organ or in organs of the same system. Of the patients with malignant neoplasms seen at the Mayo Clinic in one year, 3.4 per cent had more than one primary malignant neoplasm. Reasons have been presented to show that the percentage is even higher. From the study of cases of multiple malignant neoplasm it seems that the factors which cause the development of the tumors also express themselves in the grade of malignancy of the tumors. A large percentage of patients with multiple primary malignant neoplasm give histories indicating that other members of the family have had malignant tumors. No conclusion as to the occurrence of multiple malignant neoplasm with reference to sex could be drawn. In this series, the average age at which the growths developed was 50.4 years. Seventy-one cases of multiple primary malignant neoplasm have been described in the literature.

## AUTHORS' SUMMARY.

**SWEAT GLAND CANCER OF THE BREAST.** B. J. LEE, G. T. PACK and I. SCHARNAGEL, Surg., Gynec. & Obst. 56:975, 1933.

The human breast develops as a modified apocrine sweat gland. Apparent sweat gland tubules and cysts occur in the normal adult breast, where they anastomose with the interlobular lacteal ducts. The characteristic features which distinguish the mammary sweat gland tubules from the lacteal ducts are: constant eosinophilia of the cytoplasm, an inner layer of high columnar cells, the occasional presence of myo-epithelial cells surrounding the tubules and the tendency to form intratubular and intracystic papillary tufts. The anatomic and staining characteristics of these cells persist through all the transitional phases of normal sweat gland tubules, cysts, intracystic papillomas, adenomas and carcinomas. Evidence is presented to substantiate the theory that sweat gland carcinomas of the breast may develop from preexisting sweat gland tubules, cysts and papillary adenomas. The various stages in this transition have been seen. Except for the peculiar properties of sweat gland structures in the breast which have been enumerated, the sweat gland carcinomas of the breast have much the same structure as other mammary cancers; e. g., one finds that the bulky adenocarcinomas, the comedo-carcinomas, the papillary, intraductal and intracystic carcinomas, the medullary carcinomas, the carcinoma simplex and even scirrhouss carcinoma of the breast are

represented in this group. Sweat gland cancers of the breast occur more frequently in swarthy brunettes, whose skin has large-pored, oily, coarse texture. Their regional distribution is mostly on the periphery of the breast, particularly in the axillary tail and submammary fold. The frequency of pain, adherence of the skin and ulceration are significant clinical features of sweat gland cancer of the breast. The degree of malignancy and the prognosis following treatment are practically the same as for the general group of mammary cancers.

#### AUTHORS' SUMMARY.

### Technical

SEDIMENTATION OF THE BLOOD IN SCHIZOPHRENIA. H. FREEMAN, Arch. Neurol. & Psychiat. **30**:1298, 1933.

Freeman studied by the method of Rourke and Ernstene the sedimentation test in fifty normal persons and forty-seven carefully selected patients with schizophrenia without visceral or other organic complication. He found that in schizophrenia the sedimentation rate was not higher than in the controls, regardless of the subtypes of this mental disorder, the length of hospitalization, the degree of mental deterioration, the content of sugar, cholesterol or nonprotein nitrogen in the blood, the blood volume, the total leukocyte count and numerous other factors.

GEORGE B. HASSIN.

DIAGNOSIS OF SYPHILIS FROM A DRIED DROP OF BLOOD. ALEJANDRO CHEDIAK, Arch. de med. inf. **1**:3 and 125, 1932. P. DAHR, Deutsche med. Wchnschr. **60**: 94, 1934.

The first paper of Chediak was a preliminary report; the second contains the technical details. A drop of capillary blood from the ear or finger is obtained on a glass slide. It is immediately defibrinated by stirring and permitted to dry. The test is best made soon after the blood has dried, but delays of a few days do not interfere with the test. To the dried defibrinated blood is added 0.015 cc. of a solution containing 3.5 per cent sodium chloride and 0.3 per cent sodium carbonate. The mixture is then transferred to another slide within a paraffin ring. The commercial clearing extract for the Meinicke test (M.K.R.) and the already mentioned salt solution are heated separately at 56 C. and then mixed in the proportions of 1:10. Of the mixture, 0.03 cc. is added to the blood. The slide is then shaken for three minutes and kept for thirty minutes in the moist chamber. If the reaction is strongly positive, the reading can be made after ten minutes. When viewed with the microscope, negative tests show brown granules; positive tests, a coarse black precipitate. A comparison was carried out with the Wassermann test (Noguchi antigen), the Kahn test and the clearing test of Meinicke. In a series of 1,005 cases, the Wassermann test was positive in 88 per cent, the Kahn test in 94 per cent, the Meinicke test in 92 per cent and Chediak's test in 86 per cent. The intensity of the precipitate permits a quantitative estimation which compares well with that in the other tests. Chediak's method was checked by Dahr on 600 subjects. Of the 475 who had given negative reactions to other tests, 467 gave negative reactions to the Chediak test, and 8, or 1.7 per cent, falsely positive reactions. Of the 125 who had given positive reactions, 112, or 91 per cent, gave positive reactions to the Chediak test.

I. DAVIDSOHN.

PRELIMINARY TEST IN TRANSFUSION. O. THOMSEN, Klin. Wchnschr. **12**:1801, 1933.

Tube 1 receives six drops of serum from the recipient and one drop of a suspension of two drops of the donor's blood in ten drops of salt solution. The tube is placed in a water bath at 37 C., and if lysis occurs after from fifteen to thirty minutes the bloods are absolutely incompatible.

Tube 2 receives two drops of the recipient's serum, six drops of salt solution and one drop of the suspension of the donor's blood. In this mixture watch for agglutination.

If no lysis develops in tube 1 and no agglutination in tube 2, the donor is either of the same group as the recipient or a "universal donor." In this preliminary test it is essential that the serum of the recipient be free from all trace of lysis before the donor's blood is added.

**STAINING OF PROTEIN-LIPOID COMBINATIONS BY SUDAN III.** J. GOLDMANN,  
Virchows Arch. f. path. Anat. **290**:717, 1933.

Variations in the sudan III staining method reveal variable amounts of lipoid in the tissue. A combination of sudan III with alpha-naphthol brings to view a larger quantity of stained material than any other method. It stains the lipoid granules normally present in the polymorphonuclear leukocyte; the staining of these granules furnishes a criterion of the completeness of sudan III staining. Infiltrative lipoid as well as the "integral" lipoid of the lipoid-protein complexes of the protoplasm is stained by this method.

O. T. SCHULTZ.

**IMPROVED TECHNIC FOR THE ASHING OF TISSUE SECTIONS.** C. HACKMANN,  
Virchows Arch. f. path. Anat. **290**:749, 1933.

In Hackmann's modification of the Schultz-Brauns method for the preparation of ashed tissue sections, thin pieces of tissue are fixed for eight or ten days in formaldehyde vapor in a desiccator that contains a dish of calcium chloride. The sections are cut on the freezing microtome, those for the usual staining procedure being placed in water. The sections for ashing are placed in a dish of paraffin oil, from which they are floated to a slide to which a thin layer of gelatin has been applied. The oil is drained off, and the sections may be kept in a dust-free place until a convenient time for ashing. The heating should be gradual, and ashing should be done at a temperature of 350 and not over 400 C. Recognition of the nature and localization of the mineral salts in the ashed sections is facilitated by treating the latter with bromthymol blue, bromcresol purple and bromphenol blue.

O. T. SCHULTZ.

# Society Transactions

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## PHILADELPHIA PATHOLOGICAL SOCIETY

*Regular Meeting, Feb. 8, 1934*

MORTON McCUTCHEON, *President, in the Chair*

### GRANULOSA CELL TUMOR. HENRY K. SEELAUS.

Robert Meyer states that there are three types of ovarian tumors which exert a hormonal effect, viz., the dysgerminoma and the arrhenoblastoma, along the lines of masculinization, and the granulosa cell tumor, along the lines of feminization. This report places on record the case of a married woman, aged 74. She had had the menopause twenty years before. Fifteen years later she had a return of the menses, which recurred regularly every twenty-eight days and lasted for three or four days. Nothing unusual was noticed about the pelvis until three months before her admission to the hospital, when a large mass was detected which, at operation, was found to be a tumor of the left ovary weighing 1,622 Gm. and measuring 19 by 15 by 10 cm. Microscopic study of the solid portions of the tumor revealed the tissue to be composed of a very cellular ovarian stroma with numerous small collections of epithelial cells, in some instances forming solid clumps and in other instances arranged around the margin of a lumen-like space. The cells and their arrangement very closely resembled graafian follicles. While this type of tumor is usually classified as malignant there was, in the sections examined, nothing in the surrounding tissue to indicate definite infiltration by the type of epithelium which formed the greater portion of the growth. To date, which is more than six months since the operation, there is no evidence of recurrence and all vaginal bleeding has ceased.

### CONGENITAL HYPOPLASIA OF ONE KIDNEY ASSOCIATED WITH MALIGNANT HYPERTENSION. JOHN W. PARSONS.

A 14 year old white girl who was admitted to the hospital of the University of Pennsylvania in September, 1933, complained of severe headache, constipation, and pain in the right lower quadrant of the abdomen. She had been poorly developed all her life though mentally precocious. Two years before her admission she first noticed headaches which had recurred at irregular intervals since. Two months before admission abdominal pain suggested the diagnosis of acute appendicitis, but operation was deferred because of hypertension. On admission she was found to be markedly emaciated and to have extreme hypertension: 220 systolic and 175 diastolic. The fundus of each eye exhibited exudates and choking of the optic disk. The urine showed albumin, occasional casts and three or four white blood cells per high power field. The heart was not appreciably enlarged nor was there demonstrable peripheral arteriosclerosis. The patient's course was steadily retrogressive with headache and increasing visual disturbances leading to partial blindness. A subtemporal decompression was made in an effort to relieve the headaches but with only temporary success. The patient died four months after admission with signs indicating intracranial hemorrhage.

Autopsy revealed the right kidney to be about the size of a man's thumb, measuring 5 by 3 by 2 cm. and weighing 30 Gm. Its capsule was dense, and when stripped off it left a finely granular, dark red surface on which projected a large nodule of paler tissue which occupied the midportion of the organ. The poles of the kidney were fluctuant to palpation, and section revealed them to be dilated as by hydronephrosis, the wall being a dusky red and measuring 4 mm. in thick-

ness. The nodule in the midportion resembled a segment of normally formed, though sclerotic, renal tissue. The pelvis and the ureter were of normal size and showed no abnormality. The renal artery and vein were somewhat smaller than normal though in the usual position.

The left kidney was large, weighing 150 Gm. and measuring 10 by 6 by 3.5 cm. It was moderately sclerotic. The general structure was normal. The blood vessels, pelvis and ureter showed no abnormality. The urinary bladder was of the usual size, with slightly hypertrophied walls. The urethra was normal. The internal genitalia were infantile in size though of normal structure.

The heart was small with slight left ventricular hypertrophy. The brain was edematous and there was a massive recent hemorrhage in the temporal lobe as well as a smaller, older hemorrhage in the left putamen and the surrounding area.

Histologic examination of the left kidney revealed an occasional glomerulus containing small areas of necrosis within the capillary tuft with, otherwise, no glomerular changes. The tubules were moderately atrophic. The afferent arterioles showed marked hyaline degeneration with occasional necrotic arterioles. The small arteries showed moderate fibrous thickening.

Sections of the large nodule in the right kidney revealed changes similar to those in the left kidney. The thinned-out portions at the poles showed several peculiarities of structure. There were areas in which tubules filled with deeply eosinophilic hyaline material abounded, but in which there were no glomeruli. Other regions showed glomeruli and tubules of the highly cellular character and immature form seen in fetal kidneys while still other areas showed extensive hyalinization of closely packed small glomeruli. All of these regions were densely infiltrated by fibrous tissue. The mucosa lining the calices was hyperplastic, and the submucosa was richly infiltrated by lymphocytes and large mononuclear cells.

The changes in the left kidney and a part of the right kidney conform to the criteria for the diagnosis of malignant nephrosclerosis. Other areas in the right kidney were characteristic of congenital hypoplasia.

The case is presented to draw attention to the occurrence of juvenile malignant nephrosclerosis in a child having a congenital defect of one kidney.

#### MELANOMA WITH WIDESPREAD METASTASIS. MAX M. STRUMIA.

A white woman, 63 years old, applied Aug. 1, 1933, to her family physician for treatment of a lesion of the nail and nail bed of the index finger of the right hand, which appeared black and ulcerated (melanotic whitlow of Hutchinson). The physician fulgurated the lesion on several occasions. The patient applied for treatment at the Bryn Mawr Hospital on September 4. At this time the index finger of the right hand showed swelling of the dorsal surface of the distal phalanx, with ulceration measuring 20 by 20 by 8 mm. The nail and the nail bed were destroyed, and the borders of the ulcer were dark brown and firm. The finger was immediately removed at the basal joint, and microscopic examination of the lesion showed thickened, stratified squamous epithelium which dipped down into the subdermal structure, where epithelial pearls were present. Between the delicate strands of stratified squamous epithelium masses of cells were seen containing a large amount of melanin. A diagnosis of melanoma was made, and the patient was discharged.

Jan. 3, 1934, the patient was readmitted moribund and died in a few hours. At autopsy metastases were found in the skin, thyroid gland, mediastinal tissue and mediastinal lymph nodes, breasts, lungs and pleura, pericardium, cardiac muscle, diaphragm, surface of the peritoneum, mucosa of the ileum and colon, spleen and supplementary spleen, kidneys, liver, iliac nerve plexus, psoas muscle, bones, uterus, tubes and ovaries and also a few nodes in an old pedunculated fibroid of the uterus. The suprarenal glands weighed a little over 50 Gm. each and were entirely destroyed by pigmented tumor. The marrow of the rib as well as that of the sternum appeared completely black. Smears of marrow from the rib showed extremely few myeloid elements and nucleated red cells but a very large number

of hemohistioblasts (reticulo-endothelial cells) loaded with black pigment. This probably represented a blockage of these cells by the circulating pigment liberated by breaking down of the tumor cells. The fluid blood from the heart showed extremely few leukocytes, and very few were found in any of the vessels on section or in the various areas of inflammation following breaking down of the tumor. It is interesting that notwithstanding the complete breaking down of the suprarenal glands, the blood pressure was 220 systolic just before death and the blood sugar 35 mg. per hundred cubic centimeters of blood. The urine did not show melanotic pigment. Microscopic examination of the various organs revealed that all of the metastases were pigmented but that very few of the tumor cells were well preserved. These were always at the periphery of the nodes and occurred as polyhedral or elongated cells, lying irregularly, with large vesicular nuclei and in all cases with cytoplasm so filled with melanin as to obliterate almost completely every detail of the cell. It is of interest that the best preserved tumor cells were found in the atrophic uterus and the partly calcified fibroid of the uterus, where obviously the blood supply was very scanty.

GENERALIZED TORULOSIS ASSOCIATED WITH HODGKIN'S DISEASE. FRED D. WEIDMAN.

This article appears in full in this issue of the ARCHIVES, p. 227.

EXPERIMENTS ON TUMORS OF THE FROG. BALDUIN LUCKÉ.

In a recent communication (Lucké, Balduin: *Am. J. Cancer* **20**: 379, 1934) there has been described a cancerous disease of the common leopard frog. This disease is characterized by the occurrence of a single or multiple whitish tumor usually confined to the kidneys but in four instances of the present series extending to the extrarenal retroperitoneal tissue. The new growths vary in size from small whitish nodules to a mass weighing ten times more than the normal kidney. Microscopically the tumors have the appearance of richly cellular adenocarcinoma. Unlike human adenocarcinoma the frog's tumor rarely metastasizes, only 1 instance having been found in over 200 cases studied.

In the majority of the tumors there are present prominent acidophilic intranuclear inclusions having the same general character as the inclusions of herpes, varicella and certain other virus diseases. The inclusions have been observed only in the epithelial cells of the tumor; they do not occur in the normal renal cells or in the cells of other organs. It seems probable that the nuclear changes represent the activity of a filtrable virus, though it is not yet certain whether such a virus has a causal relation to the new growths.

Experiments in transmission of the tumors are likewise as yet indecisive. Material from 24 tumors has been inoculated by various routes into a total of 478 frogs. In the majority of the cases the inoculated cellular material (a cell suspension or small fragments of tumor) was absorbed without leading to local new growth. In a relatively small number of cases some evidence of local new growth was obtained, such as a moderate degree of enlargement of the fragments, in which microscopic examination showed mitotic figures. Fragments of this kind were found for as long as one hundred days after implantation.

While transplantation of tumors has been largely unsuccessful so far as local growth is concerned, a number of the inoculated frogs have acquired renal tumors. Indeed the most massive tumors observed have occurred in previously inoculated animals. If the neoplastic disease is caused by a virus it is to be expected that its activity under experimental conditions will be manifest in that tissue in which the lesions occur spontaneously. The relatively more frequent finding of renal tumors in the inoculated series seems therefore in harmony with the present belief adopted as a working hypothesis, that a virus is the causal agent of the tumors.

Further support may or may not be obtained when the still surviving members of the inoculated series are examined.

## NEW YORK PATHOLOGICAL SOCIETY

*Regular Meeting, Feb. 22, 1934*

WILLIAM C. VON GLAHN, President

## HEMORRHAGIC DESTRUCTION OF THE OCULOMOTOR NERVE. ALFRED PLAUT.

The patient was a man 65 years old, who presumably had always been in good health. For one year or more ptosis of the right eyelid had been present. Since the patient had a cataract in that eye not much attention was paid to the ptosis. Four weeks before admission to the hospital severe headaches, mostly in the region of the ophthalmic branch of the right trigeminus nerve, set in. Occasionally there were nausea and vomiting. In the hospital the patient's blood pressure was found to be 160 systolic and 90 diastolic, and the clinical symptoms led to a diagnosis of ruptured aneurysm of the circle of Willis. Four days before death he suddenly lost consciousness; stupor and rigidity developed. The right oculomotor nerve was completely paralyzed. The spinal fluid contained fresh blood and showed xanthochromia at the same time. It had a pressure of 320 mm. of water.

At autopsy a subarachnoid hemorrhage, not very large, was found at the base of the brain. In the region where the aneurysm had been suspected by the neurologists an aneurysm-like, bluish sac the size of a large pea was found. At first glance it was considered to be an aneurysm of the posterior branch of the circulus arteriosus. After removal of the clotted material, however, the basal arteries were found completely intact, not even sclerotic. The bloody mass was the proximal stump of the severed right oculomotor nerve. During the removal of the brain no attention was paid to the distal portion of the oculomotor nerve. The first portion of the nerve was not hemorrhagic, the hemorrhage beginning gradually. The tip of the stump was extremely thin and ruptured during dissection. Microscopically the nerve tissue could be demonstrated even at the tip of the hemorrhagic stump. In the proximal portion of the nerve mild inflammatory changes were seen. Otherwise the brain was not remarkable except for moderate chronic internal hydrocephalus.

The unusual localization of the hemorrhage and the even more unusual self-amputation of the nerve cannot be satisfactorily explained. Hemorrhage in the oculomotor nerve has been described; for instance, in tuberculous meningitis. In one of the two cases described by M. Saenger in 1880 the hemorrhage in the oculomotor nerve was the only hemorrhage found in the brain. Another similarity between that case and this one was the change in the shape of the nerve, the cross-section becoming angular instead of round.

## SUDDEN DEATH DURING LABOR. ALFRED PLAUT.

An obese, otherwise healthy, 25 year old Austrian Jewish primipara entered the hospital in the morning, the cervix partly dilated. Five hours later the cervix was nearly fully dilated and everything seemed normal, when she suddenly was taken with cyanosis, gasping and frothing at the mouth, and died within twenty minutes without convulsions. At autopsy the major portion of the pons was found occupied by hemorrhage, which had broken into the ventricles. Gross and microscopic examination of the blood vessels of the brain revealed no abnormality. The microscopic picture of the liver was normal; the kidneys showed a moderate degree of parenchymatous and hyaline change in the tubular epithelium and slight thickening of the walls of the glomerular capillaries. The antepartum course of the patient had been perfectly normal. In the last few months of her pregnancy her systolic blood pressure had risen from 108 to 125 mm. of mercury without a rise in the diastolic pressure (68). In December 1933 she had swelling of the feet, but her urine at that time was normal. A catheterized specimen taken at the onset of her fatal attack contained much albumin and occasional hyaline and granular casts.

In an attempt to find the explanation of this death, the following facts appear. The woman was very obese. Her liver was twice the normal weight (2,250 Gm.). It measured 30 by 21 by 8.5 cm. The spleen was also about twice the normal weight (330 Gm., measuring 15.5 by 9.5 by 5 cm.). In the absence of anomalies of tissue, the size of these organs must be taken as indicating a splanchnomegaly. On microscopic examination of the liver, unusually large nerve structures were noted in the surroundings of the large arterial branches. In the anterior lobe of the hypophysis a nerve structure 0.2 by 0.125 mm. was found. Such a finding is extremely unusual. Normally the hypophysis contains only very fine nerve fibrils which need special methods for demonstration. In many thousands of slides from the hypophysis I have never seen any nerve structure in the anterior lobe. The anterior lobe contained a rather large adenoma composed of pregnancy cells. In the center of the adenoma there was a hemorrhage, at the edge of which organization was seen. Thus this hemorrhage must have been older than the fatal pontile hemorrhage. The shape and staining characteristics of the hypophyseal cells outside the adenoma were very irregular.

Thus it seems that this hemorrhage occurred in a patient with multiple constitutional abnormalities, namely, abnormal, nerve structures, splanchnomegaly and obesity. In addition, there must have been a renal disturbance, as indicated by the findings in the urine. Spontaneous hemorrhage of the brain in a really normal person during labor probably never occurs.

#### ALLERGIC INFLAMMATION OF THE LUNGS. B. M. FRIED (by invitation).

The report is based on experimental observations. Rabbits were sensitized by repeated intraperitoneal injections of horse serum. While some animals were sensitized with 20 cc. of horse serum, others required 40 cc. and more. The shocking injection of 1 cc. of serum was given intratracheally about one week after the last intraperitoneal injection. The animals were killed at intervals of from one hour to several weeks after the shocking injection. Studies were made of sections cut through the entire thickness of the lungs.

On gross examination areas of consolidation and discoloration were noticed in the lungs from animals that were killed in from twelve to fifteen hours after the intratracheal shocking injection, these areas being usually confined to the base or to the paravertebral region of the lung. Within twenty-four hours after the shocking injection about 60 per cent of the animals showed an entire lobe or a whole lung to be firm, with wide hemorrhagic areas scattered through the consolidated organ.

Under the microscope, too, changes were as a rule confined to one lung, involving the interstitial and the parenchymatous elements. Granulocytes and serum flooded the septal capillaries and the alveoli, leading to their immense distention shortly after the intratracheal injection.

This stage was succeeded by another in which the intra-alveolar exudate was composed of macrophages and eosinophils. Fibrin made its appearance within a few hours after the shocking injection.

There occurred profound changes in the vessels, such as disintegration of the vascular wall, which was infiltrated with round cells. The presence of fibrin in the wall, in the lumen and also in the edematous perivascular spaces was typical of the lesion. The vessels were surrounded by collars of "monocytoid" cells. The usually "quiescent" vascular endothelium showed morphologic changes and detached itself from the wall as single cells and en bloc. In many instances there occurred complete occlusion of vessels, with thrombus formation followed by infarctions.

In brief, the studies have revealed two points of particular interest: (1) a type of pulmonary inflammation that resembled genuine or lobar pneumonia; (2) vascular changes that looked like those observed in rheumatic fever and in periarteritis nodosa. It is remarkable that lesions of such grave nature can be induced with sterile horse serum without the intervention of pathogenic micro-organisms.

## DISCUSSION

ALFRED PLAUT: Dr. Fried has mentioned the similarity of the vascular lesions to those seen in rheumatic fever and periarteritis nodosa. I must admit that I do not see that similarity. I see a simple inflammation of the blood vessels, and I wonder if other slides not depicted here really do show similarity between the allergic lesions in these lungs and the lesions found in periarteritis nodosa and rheumatic fever.

DAVID P. SEECOF: I should like to ask a few questions. How were the intratracheal injections made? Was a tracheotomy done, or was the injection made past the glottis? Does varying the dose, the amount injected into the trachea, cause any change in the reaction? In other words, was the reaction influenced by the dosage, and, if larger doses were given, were more lobes involved? Finally, how were the smaller bronchioles in the diseased lungs?

IRVING GRAEF: Were the changes confined to the consolidated lung, or were any also seen in the other lung or other organs? Also has Dr. Fried tried any species other than rabbits?

SYLVAN E. MOOLTEN: Can Dr. Fried distinguish either in degree or in kind between the inflammatory lesions he points out as allergic pneumonia and the simple aspirational reaction produced by foreign serum alone without previous sensitization or by extravasated blood? Did he find edema and fibrin in these alveoli as another evidence of an inflammatory reaction?

WILLIAM C. VON GLAHN: I should like to know whether there was fibrin in the walls of the vessels and what changes were found in the elastic tissue of the vessels.

B. M. FRIED: In answer to Dr. Plaut's question: I do believe the changes which I found resemble those seen in periarteritis nodosa and rheumatic fever. I should not say that it was a 100 per cent resemblance, but there were definite similarities between the condition which I observed in these lungs and the changes which I have seen in periarteritis nodosa and in some cases of rheumatic fever.

The second question concerned the method of injection. It was performed without a tracheotomy. The animal was placed on its back and fastened on the table; an incision was made in the skin, the muscles were separated, a sound was brought underneath the trachea and a fine needle was inserted into the trachea and the fluid pushed into the lung. It reached the finer bronchioles and the alveoli, without a doubt.

As to the influence of the dose on the reaction: I said before, serum is not an indifferent substance when injected into an animal's lung. A dose of 2 or 3 cc. produces a very definite reaction in the lung which will not subside for months, just as when one injects iodized poppy-seed oil 40 per cent or any oil it is slowly absorbed and remains for months in the lung in nodules, ultimately leading to secondary changes in the bronchi. Small amounts of serum were given in these experiments, and if the animal was properly sensitized these produced an effect. It was never necessary to inject more than 0.5 or 1 cc. I tried larger doses on normal animals, and found that with an amount of serum greater than 1 cc. the changes were permanent and grave. In these cases both lungs were involved, and the reaction was entirely different from that in these sections from allergic animals.

There was a question about the smaller bronchioles. In the early stage the reaction was not confined to the bronchioles. At a later date secondary changes in these structures occasionally occurred. Proliferation of the bronchial lining was noted, with possibly metaplastic changes.

In reply to Dr. Moolten: When a small amount of horse serum was given intratracheally there was no hemorrhage in the lungs. In these cases, the hemorrhage observed was the result of the vascular changes and of infarction.

There was considerable edema, sometimes leading to a marked distention of the alveoli.

In regard to changes in the other lung, there were none.

There were some changes in the liver and very slight changes in the heart. I was trying to be very cautious in the interpretation of my findings, and I did not want to go into details of the other organs. I therefore confined myself to the observations in the lungs.

In animals other than rabbits, such as guinea-pigs or rats, the induction of a local allergic reaction of the lung was a difficult matter. A study along this line is in progress.

Fibrin was present not only in the wall of the vessel but in the lumen and in the perivascular spaces. It is interesting to note that fibrin can be found in and around the walls of the vessels in the very early stages of the disease. I spoke of the fibrinoid reaction; actually shreds of fibrin could be seen in the wall of the vessel, in the lumen and in the perivascular spaces. The elastic tissue of the vessels was gravely injured.

#### DERMATOMYOSITIS: A REPORT OF TWO CASES. SIGMUND WILENS and ABNER WOLF (by invitation).

Two cases of dermatomyositis with the observations at autopsy are presented. One case was that of a man 53 years old, whose illness began three months before admission to the hospital with a dermatitis involving the skin of the upper extremities and face. Shortly thereafter he had weakness of the arms and legs. This was progressive and was followed by difficulty in swallowing and speech. On admission, he appeared chronically ill, showed extreme muscular weakness with considerable atrophy involving all the muscle groups, and had a dry, scaling, pruritic rash involving the face, chest and extremities. There was dysphagia. Mild fever was present; the white blood cell count was 24,000 with 85 per cent polymorphonuclears and 1 per cent eosinophils. The day following admission the patient, after finishing his lunch, suddenly became cyanotic, lapsed into stupor and died. At autopsy an extensive rash was noted over both forearms, upper arms and shoulders. The skin in these areas was scaly and roughened and slightly injected. It felt firm and was fixed to the underlying tissues. There was no pitting edema. All the skeletal muscles were pale, being light grayish red. They seemed to be normal in consistency.

The second case was that of a woman 47 years old, whose initial symptom was a dermatitis which developed eight months before admission to the hospital and involved the upper extremities. It was described as a red, scaling eruption. Three months later the dermatitis began to clear up, but eight weeks before admission difficulty in swallowing and speaking developed. Weakness in the legs and back soon appeared. On admission she appeared chronically ill and showed evidence of a loss in weight. Her speech was unintelligible, accompanied by great effort and followed by marked fatigue and dyspnea. The cutaneous eruption had entirely cleared save for some residual thickening. She showed increasing dyspnea and dysphagia and finally died. The clinical diagnosis was not definitely established, but the condition was thought to be bulbar paralysis terminating in poliomyelitis. At autopsy there were found slight scaling and thickening of the skin of the neck, hands, forearms, elbows and soles. There was no evidence of edema. The temporal, pectoralis minor and intercostal muscles were very pale, being grayish pink. The psoas major muscle, the right rectus femoris and the muscles of the back were also paler than normal, but the remainder of the skeletal muscles examined appeared grossly to be normal.

The microscopic findings in both cases were typical of dermatomyositis. No lesions were found in the central or the peripheral nervous system in either.

The pathology of the disease is as follows: The skin shows flattening of the papillae, marked increase of fibrous tissue in the cutis, edema, and mild round cell accumulations about the vessels and sweat glands. The muscles show loss of striations, fragmentation of fibrillae, hyaline or waxy degeneration, vacuolation, ruptured fibers, marked irregularity in the size and increase in the number of muscle and sarcolemma nuclei, blebbing of the sarcolemma sheath, perivascular

round cell infiltration, thickening and edema of the perimysium and endomysium and regenerative phenomena. In the more chronic cases there is a replacement fibrosis.

In the second of the cases, the creatine content of many of the affected muscles was studied. It was found to be definitely lowered, and there was a definite parallelism between the degree of involvement of a given muscle and the diminution of its creatine content. This corresponds to the findings of Steinitz and Steinfeld in the only other case of dermatomyositis in which such determinations have been made.

The etiology of the disease is unknown. Many different causative agents have been suggested. Unverricht suggested a gregarine; Langsteiner, a streptococcus; Bauer, a staphylococcus; Martinotti, a micrococcus, and others, different organisms. None of these organisms has been consistently found in the disease. Strümpell called attention to the occurrence of tuberculosis in some of the cases and thought there was a possible relationship. Grumke reported a case with a typical clinical syndrome in which the lesions were obviously tuberculous. The disease has been observed following infection of the upper respiratory tract, acute rheumatic fever, some of the acute infectious diseases of childhood and gastro-intestinal intoxications. It is possible that more than one agent may produce this condition or be predisposing factors. It is interesting that in our cases foci of infection were present: in the first case, a periappendical abscess; in the second, a tonsillar abscess. The type of onset, the fever, the angina, the frequently enlarged spleen and the type of the inflammatory changes in the muscle suggest the presence of an infectious process. Degenerative changes with secondary inflammatory phenomena in the skin and muscles due to toxins from some other primary condition form another possibility. Lesions produced in guinea-pigs and rabbits by Pappenheimer and Goettsch by special diets show some of the features of the muscular lesions of dermatomyositis. No definite evidence of improper nutrition has been encountered in the known cases of this disease, however.

#### DISCUSSION

PAUL KLEMPERER: In one of the two cases of dermatomyositis which I have seen, I observed arterial necrosis in some of the muscles, an observation which had already been made by Fahr in one of the cases which he reported. The presentation here is so detailed that it seems superfluous to ask whether any other changes were found. I presume not, otherwise they would have been mentioned, but I want to record this observation.

There is one question I want to ask: Was eosinophilia present during life, a finding which seems to be rather frequent in the cases reported? The most peculiar and characteristic feature of this disease is its unusual localization in the skin and musculature and occasionally in nerves, as mentioned, and it might be apropos to mention another case of myositis that I have seen in which the difference was that streptococci were found in great numbers in the musculature. This concerned a child who suffered with severe muscle pain for six days and died. At autopsy a large, septic spleen was found. Cultivation of this gave hemolytic streptococci in pure culture. All the muscles that were involved showed severe inflammation and necrosis, with an extremely large number of streptococci. The case may be considered as one of streptococcal septicemia with an unusual localization in the musculature. No other organ was affected. In some way this might throw light on the possible selective localization of toxins, Dr. Wilens having mentioned the infectious origin in his cases and in most of the cases cited in the literature of dermatomyositis.

ARTHUR SCHIFRIN: Was there any evidence of nephritis?

ROBERT O. FISHER: Does the disease occur without the cutaneous lesions?

ABNER WOLF: In answer to Dr. Klemperer's question: Neither case evidenced involvement of the arterial walls. Within the last week we saw a third

case in which there was marked arteritis in addition to the other changes which we noted. In neither case was an eosinophilia observed.

In reply to Dr. Schifrin's question: We did not observe any definite changes in the kidneys in either of the cases.

In answer to Dr. Fisher: The disease can occur without evident changes in the skin. The changes in the skin may be fleeting and may not be impressive enough to be noted in the clinical picture, although in the majority of cases described in the literature there have been cutaneous lesions. As Dr. Wilens observed, these skin changes may have a variable relationship to the appearance of the muscle symptoms; they may come before the muscle symptoms or they may appear very late in the disease.

#### GIANT CELL LEUKEMIA. PAUL KLEMPERER.

A case of chronic myeloid leukemia in which there were no unusual clinical, hematologic or gross anatomic features but in which the histologic examination revealed an excessive number of megakaryocytes within the spleen and lymph nodes is described. The origin of the megakaryocytes could be traced to hematic stem cells (hemocytoblasts), but occasionally the fixed reticulum cells of the spleen and lymph nodes also showed evidence of transformation into giant cells. The endothelium of the splenic sinus, however, showed no evidence of proliferation. Conspicuous accumulations of megakaryocytes and erythroblasts were found in the peripheries of anemic infarcts in the spleen.

#### DISCUSSION

MAURICE N. RICHTER: Cases of the type that Dr. Klempner has reported are very unusual, and I think they are even more unusual than he has indicated. I have seen a somewhat similar one recently which I think corresponded a little more to the type usually described in the literature. Many cases in which the megakaryocytes have been unusually common have been cases of aleukemic myelosis. Furthermore, a number of the cases have been associated with osteosclerosis, and in the one which I studied osteosclerosis was present in the sternum, femur, ribs and vertebral marrow. The sclerosis was present to such an extent that the remaining marrow could not be recognized as that from a patient with myeloid leukemia, and megakaryocytes in particular were practically absent from the bone marrow, so that one can hardly consider them as embolic from that source. Cases in which osteosclerosis, a low white blood cell count and numerous megakaryocytes were present have been observed a number of times, so often that it seems to me there must be some significance in the association of these three rather unusual features.

ALFRED PLAUT: In January I had an opportunity of observing something similar in a patient who had had polycythemia. A year before death this patient had 16 liters of blood. Then the polycythemia disappeared, and the patient died with old abscesses of the liver and many other lesions. The first thing that drew attention to the fact that the patient had an aleukemic leukemia was the large number of megakaryocytes in the lymph nodes, spleen and liver. This was not considered to indicate a special disease, but to be just an observation of aleukemic leukemia with a large number of megakaryocytes, and there was no hesitation in assuming that the megakaryocytes had originated in the spleen and in the lymph nodes. As to the question of origin, one might cite the simple fact that certain animals have megakaryocytes normally in the spleen, and probably nobody would think of them as of embolic origin.

SEATON SAILER: I should like to ask if there is any explanation for the reduction in the number of platelets, in view of the proliferation of the megakaryocytes and the fragmentation of some of them.

PAUL KLEMPERER: The fact that in osteosclerotic leukemia the megakaryocytes in the internal organs are increased in number was also borne out by one of the other cases in my series. There was also leukopenia.

Regarding Dr. Sailer's question as to the explanation for the low platelet count in this instance, I can refer to other cases in which the same observation was made. It is peculiar that in spite of the marked proliferation of megakaryocytes and the marked fragmentation there were not more megakaryocytes in the circulating blood. There is one factor that has to be considered, namely, that the presence of megakaryocytes and the separation of platelets within the spleen need not lead to an increase in the number of platelets in the general circulation, for they might be destroyed within the portal system and then might be filtered out in the lung. Another explanation has been given, that the megakaryocytes do not properly form platelets. In the case which I have described this was not suggested, for one could see the formation of platelets from the megakaryocytes within the tissues.

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## CHICAGO PATHOLOGICAL SOCIETY

*Regular Monthly Meeting, Feb. 12, 1934*

E. H. HATTON, *President, in the Chair*

### INFECTIOUS MONONUCLEOSIS. I. DAVIDSOHN.

The demonstration by Paul and Bunnel of an increase in heterophilic antibodies in the blood of patients with infectious mononucleosis is an important contribution to knowledge of the disease. This observation has been confirmed repeatedly. My report is presented to emphasize the value of an examination of the serum for heterophilic antibodies.

A 19 year old white boy, a patient of Dr. Richard Gordon, was admitted to the Mount Sinai Hospital, Sept. 28, 1933, with pain in the abdomen and in the right axilla which had lasted for five days, and a swelling in the right axilla of three days' duration. At first the pain in the abdomen was generalized; then it localized in the lower portions. He had a feeling of distention but no nausea or vomiting. Constipation of three days' duration was relieved by a cathartic. His past and family histories are not important. Physical examination revealed that the cervical, left axillary, and both inguinal lymph nodes were enlarged. In the right axilla was a mass about the size of a walnut. The nodes were tender, the mass in the right axilla more than the others. The firm, tender spleen extended about 1 fingerbreadth below the costal margin. The liver was enlarged and tender. There was some sensitiveness in the right lower quadrant of the abdomen. The temperature on admission was 100.8 F.; the pulse rate, 88; the respiratory rate, 24. Acute appendicitis was considered. Examination of the blood on the following day revealed a leukocytosis of 13,750 in which 2 per cent were band forms. 12 per cent segmented neutrophils, 1 per cent eosinophils and 1 per cent basophils. The remaining 84 per cent were mononuclear cells, of which 63 per cent were lymphocytes, 1 per cent plasma cells, 6 per cent monocytes and 14 per cent abnormal lymphocytes. The conditions in the blood and the clinical symptoms suggested infectious mononucleosis. The test for heterophilic antibodies in the serum demonstrated a high titer (1:5,120) of agglutinins for sheep red cells. (The technic of the test will be presented in detail in an extended report.) Sections of a lymph node from the groin showed hyperplasia of the endothelial cells. The tissue changes were similar to those described in previous reports. The patient left the hospital on the third day after admission but remained under observation. The spleen was not palpable on the eighteenth day after the onset of symptoms; the lymph nodes were not tender, but their size decreased very slowly, and some were distinctly enlarged after four and a half months. Abnormal lymphocytes were seen until three weeks after the onset, but the mononucleosis (51 per cent) was present after four and a half months.

COALESCED GLANDULAR CARCINOMA AND LYMPHOSARCOMA OF THE COLON.  
C. S. HAGERTY.

Malignant tumors of both carcinoma and sarcoma are rare. Glaessen and Mathias (*Beitr. z. path. Anat. u. z. allg. Path.* **123**:584, 1921) compiled seventy-one reports of such tumors and Jaffé (*Surg., Gynec. & Obst.* **37**:472, 1923) thirty-two. Ehrlich and Apolant (Ewing, J.: *Neoplastic Diseases*, ed. 3, Philadelphia, W. B. Saunders Company, 1928, p. 1018) obtained a tumor with epiblastic and mesoblastic tissues by transplanting carcinoma through several generations of mice. In man, tumors of sarcoma and carcinoma occur with greatest frequency in the uterus, ovary, thyroid gland and digestive tract. Schubach (*Ztschr. f. Krebsforsch.* **33**:126, 1931) described a gastric tumor of undifferentiated carcinoma and lymphosarcoma. A tumor of the colon, similar in structure to that described by Schubach but with more clearly differentiated tissue components, is now reported.

A white woman, aged 59, a patient of Dr. L. E. Schmidt, entered St. Luke's Hospital, June 20, 1933, because of pain in the left kidney of eight months' duration. She had had pulmonary tuberculosis in 1925; in 1931, the uterus was removed because of a glandular carcinoma of the fundus. A parent and a sister had died of carcinoma. A pyelogram of the left kidney demonstrated a nearby mass which by operation was determined to be a tumor of the descending colon. The excision of the growth was done by Dr. C. G. Shearon. The patient did not survive the operation.

The colonic segment was 12 cm. long and 9 cm. in circumference. In the center of the lining was an annular ulcerated growth with rounded edges, 3.5 cm. in diameter, projecting 4 cm. above the mucosa. The cut surfaces were opaque gray-white. The lining surface covering the tumor was uneven; in the ulcerated center was a stellate perforation 3 mm. in diameter. The mucosa about the tumor was unchanged.

In the histologic preparations large regions of necrosis were found. There were two types of tissue, varying in amount, in the places examined. The first, mainly along the lining edge, was glandular carcinoma in tubules and papillae. The stroma consisted of fibroblastic tissues with inflammatory exudates of the second variety of tumor tissue. This had a delicate fibrillar stroma with the polymorphous cells and grouping seen in the Kundrat lymphosarcoma.

The postmortem examination, two hours after death, showed: surgical anastomosis of the colon below the splenic flexure (resected neoplasm); edema of both lungs; chronic fibrous myocarditis, and chronic fibrocalcareous tuberculosis of the lungs. The examination was restricted to the thorax and abdomen, in which careful search failed to reveal metastases in the viscera.

Tumors of malignant epiblastic and mesoblastic tissues, as a rule, are designated carcinosarcoma or sarcocarcinoma, depending on the type of tissue predominant. Gotting (*Frankfurt. Ztschr. f. Path.* **41**:107, 1931) stated that a carcinosarcoma may arise in one of the following ways: (1) the stroma of a carcinoma may become a sarcoma; (2) a carcinoma and a sarcoma may develop separately and as a result of rapid growth show an intermingling of their cells, or (3) carcinoma and sarcoma may develop from a matrix having epiblastic and mesoblastic potentialities. A fourth, but now discredited, possibility is the transformation of a carcinoma into a sarcoma, or vice versa.

The carcinosarcoma produced experimentally in animals is thought by Coenen and Simmonds to demonstrate that the stroma of a carcinoma has developed into a sarcoma. They refer to this growth as a mutation tumor, to designate a change in the quality of the tissue. The implanted tissue is usually carcinoma, and after transplantation through several generations of animals the pure carcinoma may change to carcinoma and sarcoma. The stimulating factor of the carcinoma, it is thought, is responsible for this change. The rarity of carcinosarcoma, however, seems to preclude this possibility.

Two malignant tumors may develop in the same organ independently. Because of their close proximity and rapid growth the two types of malignant tissue intermingle, and the resultant tissue is that of a carcinosarcoma. (collision tumor). The tumor elements are related to each other as stroma is to parenchyma. Tumors of this type are usually found in the uterus and the ovaries.

The tumor of the colon described is regarded as a glandular carcinoma and a lymphosarcoma developing independently, though possibly due to a common stimulus. The close proximity of the two tumors resulted in fusion. If named according to the possibilities of origin, the tumor is properly called a collision carcinolymphosarcoma. Schubach interpreted the tumor in his patient similarly and designated the growth a collision carcinolymphosarcoma of the stomach.

Apparently the tissues of my patient had some inherent tendency to become malignant, since a carcinoma of the uterus and a combined glandular carcinoma and lymphosarcoma of the colon developed within two years.

#### DISCUSSION

EMIL RIES: Years ago a search was made for sarcoma of the endometrium in the presence of carcinoma of the uterus. No attention is given now to this possibility.

P. DELANEY: Malignant tumor of the thyroid gland with spindle-shaped cells has been considered a mixed tumor, but I doubt that it is, because epithelium can behave very strangely in tumor tissues.

E. F. HIRSCH: A malignant growth of the thyroid gland (*Am. J. Cancer* 15:55, 1931) had long spindle-shaped cells with the cross-striations of skeletal muscle. Even remote metastases had cells with these characteristics.

#### PURIFICATION OF THE VIRUS OF POLIOMYELITIS. JAMES A. HARRISON.

The virus of experimental poliomyelitis has been treated in a variety of ways designed to effect purification. My associates and I have had occasion to use the method of Clifton, Schultz and Gebhardt, employing treatment with ether, and the method of Sabin, using aluminum hydroxide adsorption, in the preparation of virus for serologic studies. A combination of these treatments has enabled us to obtain from emulsions of the cord tissue of infected monkeys water-clear, colorless virus preparations which have been uniformly infectious in a large number of tests. The virus present in concentrates of such preparations (produced by boiling them in vacuo) survives at least four hours of tryptic digestion and subsequent dialysis against distilled water for twenty-four hours. These final virus preparations have not been anticomplementary in comparative 40 per cent suspensions, whereas crude suspensions of the virus-containing cord tissue are anticomplementary in 1 per cent suspensions.

#### DISCUSSION

W. J. NUNGERSTER: There is a possibility of loss of virus through these manipulations. The loss probably would be less with desiccation in vacuo than in the presence of oxygen.

#### LEIOMYOSARCOMA OF THE ADULT KIDNEY. P. A. DELANEY.

Oct. 15, 1933, a white man, aged 51, had an acute pain in the region of the left ilium radiating to the left lumbar region. A tender, hard mass, about the size of a large grapefruit, freely movable, thought to be the left kidney, was palpable in the left side of the upper part of the abdomen. He had had three intermittent attacks of hematuria during fourteen months; during the past six months he had lost 40 pounds (18.1 Kg.) in weight and had been lethargic, and for four months he had had no appetite. October 16, the patient entered the Englewood Hospital. Roentgen examination demonstrated a markedly enlarged left kidney, which was removed, October 21. It weighed 480 Gm. The outer surface was

nodular and hemorrhagic. Friable tissues perforated the capsule laterally, into the pelvis and hilus, and extended along the renal vein. The capsule stripped readily. A frontal section revealed friable, hemorrhagic tumor tissue and small masses of renal cortex and medulla. There were tumor thrombi in the main portion and in the large branches of the renal vein. Microscopically, no part of the kidney was unchanged. The cortex and medulla were replaced by a tissue morphologically and tinctorially spindle-shaped smooth muscle cells with intracellular fibrillae. Considerable tumor tissue was necrotic. Small masses of renal parenchyma, not invaded by the tumor, had glomeruli and tubules distorted by pressure. There were many casts in the dilated tubules. Portions of the renal pelvis and of the upper part of the ureter were invaded by tumor tissues. Large and small veins were completely occluded by tumor thrombi. Myoblasts infiltrated the fat tissues of the renal hilus.

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*Regular Monthly Meeting, March 12, 1934*

E. H. HATTON, *President, in the Chair*

OSTEOMALACIA IN A MAN. FRANCIS D. GUNN and WALTER H. NADLER.

A clinical description of this case was published in 1917 by Elliott and Nadler (*Tr. Chicago Soc. Int. Med.* 1-2:56, 1917). Evidences of the disease appeared when the patient was about 19 years of age and continued for twenty-one years. During eleven years of observation no significant findings other than the changes in the bones were made. There was no history of dietary deficiency. Castration apparently did not alter the course of the disease. Pathologic fractures occurred late in the disease, and death occurred when the patient was 40 years of age, following an acute infection of the upper respiratory tract and cardiac decompensation.

Necropsy revealed fatty degeneration of the myocardium, chronic passive hyperemia of the liver, parenchymatous degeneration of the kidneys and generalized decalcification and rarefaction of the bones without the presence of cysts or brown tumors.

The glands of internal secretion were normal grossly and microscopically with the exception of the hypophysis and parathyroid glands. The hypophysis showed slight hypertrophy (0.95 Gm.), but without selective hyperplasia of any one type of cell. There were only two parathyroid glands; each was definitely enlarged, but microscopically the only deviation from normal was a diminished number of fat cells and mild simple hyperplasia of the parenchyma. The renal stroma contained numerous small calcareous deposits.

The hypertrophy and hyperplasia of the parathyroid glands are interpreted as compensatory, due to increased physiologic activity.

The report will be published in full in the *Archives of Internal Medicine*.

METEOROLOGICAL FACTORS IN THE EPIDEMIC OF POLIOMYELITIS. WILLIAM F. PETERSEN.

Evidence was first presented concerning the association of a severe polar infall with the precipitation of a syphilitic poliomyelitis. The city records indicate precipitation of poliomyelitis at the same time. The poliomyelitic records of Detroit, Windsor, Ont., Toronto, Chicago, Milwaukee, Duluth, Minn., St. Paul, Kansas City, Mo., and other adjacent cities indicate the frequent precipitation of acute poliomyelitic episodes with the passage of polar fronts. As a result coincident cases are evident in remote cities at the same time.

Attention was called to the fact that a pressor episode (with periods of relative anoxemia in vessels of the spinal cord) may so condition definite regions of the central nervous system that localization of a virus may result in seemingly selective foci.

**"BRENNER TUMOR" OF THE OVARY OF UNUSUAL SIZE. MARSHALL DAVISON and BENJAMIN H. NEIMAN.**

An ovarian tumor weighing 15 pounds (6.8 Kg.) was found at operation in a nulliparous Negress 45 years of age. Her menses had stopped two years previous to her entrance into the hospital. The tumor occupied the position of the left ovary. The remaining adnexae uteri had no gross changes.

Histologic preparations of the tumor contained groups of epithelial cells separated by a dense stroma of connective tissue. Some of the fibrils, especially about the epithelial cells, were hyalinized. Many of the cell masses were solid aggregates; others had clefts which formed small cysts filled with mucoid material.

Foci of Walthard cells may develop in one direction or another, depending on various influences, and form abnormal structures. If the foci of Walthard cells retain their indifferent character in the course of development, the solid Brenner tumor develops. If the differentiation of the cells tends primarily in the direction of cyst formation, the second type of Brenner tumor develops in which the cyst formation reaches such an extent that a cystoma appears, with an intramural Brenner nodule.

Since "Brenner tumor" is a misnomer from a historical point of view, and is of little value in describing the condition, the term "benign mucinous or pseudo-mucinous fibro-epithelioma," suggested by Plaut, seems to be more applicable.

**DISCUSSION**

J. I. BREWER: In two patients with these growths whom Dr. H. O. Jones and I have had under observation (*Am. J. Obst. & Gynec.* **25**:505, 1933) there were manifestations of hormonal activity.

**FIBROMYOMA OF THE ILEUM. I. PILOT and SAM BROCK.**

Benign tumor of the small bowel is comparatively rare and seldom recognized before operation. We report a case because of the complication of the tumor by infection. The literature on benign tumors of the ileum down to 1927 has been summarized by Clifton and Landry (*Boston M. & S. J.* **197**:8, 1927). Other reports on these tumors are by Key Oberg (*Acta chir. Scandinav.* **62**:261, 1927), Goldsmith (*ibid.* **62**:261, 1927), Boutreau-Roussel and Cadenat (*Bull. et méém. Soc. nat. de chir.* **53**:921, 1927), Sorkness (*Journal-Lancet* **48**:146, 1928), Steindl (*Wien. med. Wchnschr.* **79**:1256, 1929) and Marintschik (*Zentralbl. f. Chir.* **56**:1362, 1929).

A white man, aged 50, a sheet-metal worker, entered the Edgewater Hospital, Dec. 18, 1930, with sharp, lancinating pain in the lower part of the abdomen, which began suddenly about thirty-six hours before. The patient said that he had had occasional similar attacks for about three months. He had had no chills or fever and had noticed no change in the character of his stools. His appetite had been poor, and he had lost about 10 pounds (4.5 Kg.) in weight; he was a small, thin man. The abdomen was moderately tender and rigid, especially over the suprapubic region. No masses were palpable. Under spinal anesthesia a right pararectus incision was made. The peritoneum contained a small amount of cloudy fluid with a foul odor. There was a diffuse peritonitis. The appendix was bound by adhesions. The pelvis contained a soft, boggy mass—a tumor attached to the ileum. It had prolapsed into the pelvis and was adherent to the rectum and sigmoid. On freeing the mass and the small bowel, the tumor ruptured, and a small amount of putrid material escaped. The tumor arose from the lateral surface of the bowel. It was soft and contained regions of fluctuation. As it was considered a malignant growth, it was resected, with 5 cm. of bowel extending proximally and distally.

The resected portion was 13 cm. long. At its free margin was a pedunculated mass, 7 cm. long and 6 cm. in diameter, that arose from the muscularis. The

pedicle was 3 cm. in diameter. A gray area of softening on the serosa extended into an abscess in the center, which was filled with foul green exudate. The remainder of the tumor was soft and grayish yellow. The mucosa of the intestine was smooth except for a small opening that led from the lumen into the abscess of the tumor. The exudate in the abscess contained many gram-negative bacilli and gram-positive cocci.

Microscopically the tumor showed interlacing bundles of smooth muscle cells and small amounts of fibrous tissue. Blood vessels were few.

The growth had several interesting features. The first clinical manifestation was an acute peritonitis, undoubtedly the result of a perforation of the abscess into the peritoneal cavity. The central abscess communicated with the intestinal lumen through a narrow fistula, with insufficient drainage, causing progressive softening and extension to the peritoneum. The tumor was outside the main plane of the intestinal wall and produced no obstructive symptoms.

#### DISCUSSION

PAUL CANNON: Recently, during postmortem examination of the body of a man, aged 45, I found in the jejunum a fleshy mass with an abscess. Histologically the tissues were like those of the tumor described by Dr. Pilot. There were secondary abscesses of the liver. Colon bacilli and streptococci were isolated from the infected tissues.

E. B. FINK: A girl, aged 6, had abdominal symptoms due to a fibroma protruding into the lumen of the ileum.

## Book Reviews

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**Précis de microscopie: technique, expérimentation, diagnostic.** By M. Langeron, Chef de laboratoire à la Faculté de Médecine de Paris, Directeur-adjoint à l'Ecole pratique des Hautes-Etudes, Secrétaire général des Annales de parasitologie humaine et comparée. Fifth edition, entirely rewritten. Price, 100 francs. Pp. 1,205, with 365 figures. Paris: Masson & Cie, 1934.

The fact that the book is in its fifth edition since it was first published in 1913 proves that it fills a need in the French medical literature. The reason for that success becomes apparent even after a casual study of its contents. The physics of the microscope, its various applications and its proper care are presented in 273 pages. This chapter is up to date and includes all recent developments in this rapidly growing technical field. A discussion of the "ultropaque" lens, which was only recently put on the market by the optical firm of Leitz, may serve as an example of the completeness of the presentation. The chapter on microphotography has some good practical suggestions. The second part, consisting of 381 pages, deals with histologic technic in its broad meaning. The beginner is introduced in a clear and effortless manner into all phases of histologic technic, but even the most expert and experienced worker will find a veritable mine of facts. Discussions of the various applications of carmine (6 pages), of hematoxylin (14 pages) and of impregnation with metals (30 pages) are picked at random as a few representative examples of Langeron's excellent presentation. Many valuable suggestions will be particularly appreciated by those who have made histologic preparations and still more so by those who have been frequently baffled by the imperfections in the slides prepared for them by technicians. They will find on page 632, in addition to many references in the text, an instructive discussion of the common errors in histologic technic and of the ways to avoid them. Each procedure is preceded by a lucid presentation of the underlying theoretical background, so that the book is not merely a compilation of technical details. The evaluation of technical methods and their pros and cons are discussed frankly and objectively. The third part deals with the study of the Protozoa and the Metazoa. It includes brief chapters on the blood, cerebrospinal fluid, sputum, milk and urine. Medicolegal microscopy is discussed on 6 pages. Sixty-five pages are devoted to bacteriologic and mycologic technic. A presentation of essentials of botanical technic and tables of physical and chemical constants complete the contents. The index and bibliographic references are very complete.

The book can be well recommended primarily for those interested in histology and in clinical pathology.

**Brucella Infections in Animals and Man. Methods of Laboratory Diagnosis.** I. Forest Huddleson, Department of Bacteriology and Hygiene, Michigan State College. Price, \$2.25. Pp. 125, with 24 illustrations. New York: The Commonwealth Fund, 1934.

This book emphasizes the common interests of the workers in human and veterinary aspects of Brucella infections. It is restricted almost entirely to a description of technical laboratory procedures for the diagnosis of Brucella infection. There are some brief references to interpretations and findings. The procedures which originated through the author's own work are presented clearly. An extensive bibliography is included. The book is a valuable technical guide; pathologists will be interested particularly in the rather short chapter on structural changes in Brucella infections.

## Books Received

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COLLECTED REPRINTS FROM THE LABORATORIES OF THE MOUNT SINAI HOSPITAL, NEW YORK. Louis Gross, M.D., Director, 1933.

URINARY ANALYSIS AND DIAGNOSIS BY MICROSCOPICAL AND CHEMICAL EXAMINATION. Louis Heitzmann, M.D., New York, Formerly Professor of Pathology and Bacteriology, Fordham University School of Medicine, New York. With a Chapter on the Determination of the Functional Efficiency of the Kidneys by Walter T. Dannreuther, M.D., F.A.C.S., Professor of Gynecology and Director of Department, New York Post-Graduate Medical School and Hospital, Columbia University. Sixth revised edition. Price, \$5. Pp. 385, with 131 illustrations. Baltimore: William Wood & Company, 1934.

The first edition of this book was published in 1899. The present edition has been brought up to date, but the chemical tests described are such as "can be performed without the necessity of a completely equipped chemical laboratory, by the technician or the physician. . . ." A short new chapter on the hormone tests for pregnancy is introduced. The illustrations, all more or less diagrammatic and rather crude, appear to be the same as those in earlier editions.

BRUCELLOSIS: A PUBLIC HEALTH PROBLEM. Ward Giltner. Pp. 118. East Lansing: Michigan State College, 1934.

THE MEDICOLEGAL NECROPSY. A Symposium held at the Twelfth Annual Convention of the American Society of Clinical Pathologists at Milwaukee, June 9, 1933. Edited for the Society by Thomas B. Magath. Price, \$2.50. Pp. 167. Baltimore: Williams & Wilkins Company, 1934.

The contents of the book are: introduction, Frederic E. Sondern; the medicolegal system of the United States, Oscar T. Schultz; the medicolegal necropsy, Charles Norris; performing the medicolegal necropsy, A. V. St. George; pathologic anatomy of death by drowning, Edward L. Miloslavich; toxicology in the medicolegal autopsy, Alexander O. Gettler; medical examiner's findings in deaths from shooting, stabbing, cutting and asphyxia, Harrison S. Martland; report on necropsies (reprinted from the ARCHIVES [14:701, 1932]) by joint committee representing the New York Academy of Medicine, the New York Pathological Society and the Metropolitan Funeral Directors' Association. These articles are reprinted from the January 1934 issue of the *American Journal of Clinical Pathology*.

REPORT OF THE LABORATORY AND MUSEUM OF COMPARATIVE PATHOLOGY OF THE ZOOLOGICAL SOCIETY OF PHILADELPHIA. Herbert Fox, M.D., Pathologist. Pp. 55. 1934.

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### CORRECTION

In the first line of the fourth paragraph of the abstract of the article by Dr. Raymond S. Rosedale, entitled "Congenital Heart Disease: Interventricular Septal Defect, Dextroposition of Aorta, and Dilatation of Pulmonary Artery," which appeared on page 721 of the May 1934 issue of the ARCHIVES OF PATHOLOGY, in the proceedings of the Buffalo Pathological Society, the word "not" should be eliminated, making the sentence read: "The normal equal division of the truncus and bulbus arteriosus had taken place, for the remainder of the aorta which is not concerned with the septum trunci was of the same caliber as the ascending portion."

# ARCHIVES OF PATHOLOGY

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## THE DIGITAL VASCULAR SYSTEM

WITH REFERENCE TO THE STATE OF GLOMUS IN INFLAMMATION,  
ARTERIOSCLEROTIC GANGRENE, DIABETIC GANGRENE, THROMBO-  
ANGIITIS OBLITERANS AND SUPERNUMERARY DIGITS IN MAN

NICHOLAS W. POPOFF, M.D.

ROCHESTER, N. Y.

In 1924 Masson<sup>1a</sup> published a paper on the subject of peculiar small subungual tumors, benign in structure and clinically manifested by much pain. He considered these tumors as being genetically related to peripheral arteriovenous anastomoses. Owing to their resemblance to the gland of Luschka, or the glomus coccygeum, Masson chose for these tumors the name glomic tumors.

Knowledge of the relation of glomic tumors to peripheral arteriovenous anastomoses involves consideration of present knowledge of the anatomy and physiology of these anastomoses. Sucquet<sup>2</sup> described peculiar vessels in several regions in the palm and sole, especially in the median zone on the ventral surface, which appeared to be larger than ordinary capillaries and which opened directly into the neighboring veins without forming a capillary network. In his studies Sucquet employed a method of injection, dissection and examination by a magnifying glass. To Hoyer<sup>3</sup> must be given the main credit for masterly anatomic studies on arteriovenous anastomoses. He used better methods than those of Sucquet, and his studies present a thorough anatomic and topographic description of arteriovenous anastomoses. The correctness of Sucquet-Hoyer's anatomic concept was corroborated later by histologic studies (Grosser,<sup>4</sup> Schumacher,<sup>5</sup> Masson,<sup>1</sup> Clara<sup>6</sup>

From the Laboratoire d'anatomie pathologique, Université de Montreal, Canada, and the Department of Pathology, the Highland Hospital of Rochester.

The work was done with the aid of the National Academy of Sciences of the United States of America.

1. Masson, P.: (a) Lyon chir. **21**:257, 1924; (b) Arch. per le sc. med. **50**:1, 1927.

2. Sucquet, quoted by Testut, I. L., and Testut, J.: *Traité d'anatomie humaine*, Paris, Gaston Doin & Cie, 1921, vol. 2, p. 101.

3. Hoyer, H.: Arch. f. mikr. Anat. **13**:603, 1887.

4. Grosser, O.: Arch. f. mikr. Anat. **60**:191, 1902.

5. Schumacher, S.: Arch. f. mikr. Anat. **87**:309, 1916.

6. Clara, M.: Ergebn. d. Anat. u. Entwicklungs gesch. **27**:246, 1927.

7. (a) Grant, R.: Heart **15**:281, 1931. (b) Grant R., and Bland, E.: ibid. **15**:385, 1931.

and others). Grant and Bland<sup>7</sup> were the first physiologists to study arteriovenous anastomoses with the use of modern physiologic methods. As to observations on the changes of arteriovenous anastomoses in pathologic conditions, nothing is found in the literature, with the exception of Masson's discussion of glomic tumors. Even Buerger's<sup>8</sup> fundamental clinical treatise on disorders of the vascular system fails to mention peripheral arteriovenous anastomoses and their possible relation to some of the peripheral vascular disturbances.

#### MATERIAL AND METHODS

Guided by the experience of the aforementioned authors, who studied arteriovenous anastomoses in various animals and in different parts of the body, I limited my investigations to the study of digital arteriovenous anastomoses in man. Cases of normal persons of different ages, including the human fetus of from 4½ months to term, were studied. My pathologic material covered the following conditions: digital cellulitis, arteriosclerotic gangrene, diabetic gangrene and thrombo-angiitis obliterans. I also had an exceptional opportunity to examine slides from Cummins'<sup>9</sup> work on spontaneous amputation of supernumerary digits (pedunculated postminimi) in man.

Almost all the previous investigators who studied the arteriovenous anastomoses employed the method of intravascular injections with coloring substances. My investigations were done with fresh surgical material fixed in situ, without being subjected to intravascular injections of fixing fluids and coloring substances. Picroformol of Bouin<sup>9a</sup> was employed as the principal fixative. In each case the pieces for embedding in paraffin were taken from the following regions: the pad (ventral surface), both lateral surfaces, the nail bed and the nail matrix. The methods of staining employed were: Masson's trichrome stain,<sup>9b</sup> Weigert and

8. Buerger, L.: The Circulatory Disturbances of the Extremities, Philadelphia, W. B. Saunders Company, 1924.

9. Cummins, H.: Am. J. Anat. **51**:381, 1932.

9a. The formula for picroformol of Bouin is as follows: formaldehyde solution, 10 per cent; water, 30 per cent, and glacial acetic acid, 2 per cent, with trinitrophenol added to saturation.

9b. Masson's trichrome stain is composed of iron, hematoxylin, fuchsin, Poinceau de xylidine (an azo dye of the aminobenzidine sulphonatronic acid group) and aniline blue. The procedure of this method is as follows: Mordant sections are immersed in iron and aluminum at 40 C. for five minutes and in iron and hematoxylin at 40 C. for five minutes. This is followed by stirring in a jar of 95 per cent alcohol for a few minutes and by differentiation in picric alcohol. The sections are then washed in running water for fifteen minutes, stained with Poinceau-fuchsin solution (fuchsin acid, 1: 100, one part, and Poinceau de xylidine, 1: 100, two parts) for five minutes and rinsed in distilled water. They are then immersed in phosphomolybdic acid for five minutes and rinsed again in distilled water. About 10 drops of aniline blue is put on the slide for from ten to fifteen minutes, and the sections are placed in phosphomolybdic acid for five minutes and then in acetic acid solution, 1: 50 or 1 per cent, for from two to three minutes. This is followed by dehydration with absolute alcohol and clearing in toluene. The sections are mounted in balsam to which 0.5 per cent of dry powdered salicylic acid is added.

Duval's stain for elastin, Mallory's stains for fibrin and silver impregnation methods of Rogers,<sup>9c</sup> Foot<sup>9d</sup> and Schultze-Stohr.<sup>9e</sup>

Digital arteriovenous anastomoses are situated in the deeper layer of the corium or in the stratum reticulare. There they occupy a narrow zone which lies parallel to the surface of the skin. For this topographic reason all the blocks of my normal and pathologic material were sectioned parallel to the surface of the skin. Such orientation permits examination of the various zones of the skin in their topographic sequence. For making quantitative studies and for measuring the individual arteriovenous anastomoses, the advantage of this method is obvious. The observations reported here are based chiefly on the method of reconstruction from serial sections.

#### NORMAL DIGITAL ARTERIOVENOUS ANASTOMOSES OF THE SUCQUET-HOYER TYPE

Before presenting my observations in cases of normal adults and fetuses, it is appropriate to summarize the essentials concerning the present knowledge of the anatomy and histology of normal digital arteriovenous anastomoses of the Sucquet-Hoyer type. As a general rule, the arteriovenous anastomoses are seen almost exclusively over the ventral surface of the hand and foot. They are constantly present in the region of the nail bed and in the tips of the digits, the palmar surfaces of the first, second and third phalanges and the thenar and hypothenar eminences of the hand. Arteriovenous anastomoses in the foot are distributed in a way similar to those of the hand. Besides being seen in the nail bed and the lateral surfaces and pads of the

9c. The method of Rogers consists in fixation in from 10 to 20 per cent formaldehyde or Bouin's picro-aceto-formaldehyde, embedding in paraffin, the cutting of sections impregnated with 40 per cent silver nitrate solution, reducing with di-ammoniacal solution toning with gold chloride solution, and fixing in 5 per cent sodium hyposulphite. The specimens are then washed, dehydrated and mounted.

9d. The method of Foot is as follows: fixation in Carnoy's solution (absolute alcohol, six parts; chloroform, three parts, and glacial acetic acid, one part); embedding in paraffin; pretreatment with a mixture of pyridine, two parts, and glycerol, one part; washing with 95 per cent alcohol; washing with distilled water; impregnation with 10 per cent aqueous silver nitrate at 37 C.; development for twenty minutes in 5 per cent neutral formaldehyde solution; developing with 0.5 per cent pyrogallol; washing; toning with gold chloride solution; intensifying by means of immersion for five minutes in oxalic acid; fixing for five minutes in 5 per cent aqueous solution of sodium thiosulphate; washing at the tap; dehydrating; running through xylene, and mounting in Canadian balsam.

9e. Variation 9 of the method of Schultze-Stohr for staining peripheral nerves consists in: fixation in 10 per cent formaldehyde; the making of a frozen section; immersion in 10 per cent sodium hydroxide for twenty-four hours; washing with distilled water for from one to two hours, with from four to six changes; immersion in silver nitrate, 10 per cent, for twenty-four hours; reduction with hydrochinon formaldehyde; dehydration, and mounting.

toes, they are found in the sole near the heel. Being formed by branches of preterminal subcutaneous arteries, the zone of the arteriovenous anastomoses is located a little deeper than the web of subpapillary arteries and veins. The topographic relation of subcutaneous arteries to branches which form arteriovenous anastomoses shows considerable variation. The main type of anastomosis is usually seen in the pulp of the finger and is formed as follows: The afferent artery arises from branches which run in subcutaneous tissue parallel to the surface of the skin. These large branches go upward and toward the surface. After they reach the inner zone of the stratum reticulare, each divides into two branches. The stronger branch bends at a right angle and continues parallel to the skin, and the weaker branch divides again. Some of the divisions of the weaker branch contribute to the formation of arteriovenous anastomoses; some go to the papillary bodies, and the artery itself ends as an artery of the papillary body. The arterial branches which become transformed into arteriovenous anastomoses acquire a peculiar appearance, owing to specific changes in their walls. The anastomosis is S-like in appearance, twisting, coiling and seldom straight. The lumen is narrow and irregular. Its endothelial cells are voluminous and cuboidal and are arranged in two or three rows; their nuclei stain deeply. There is no internal elastic lamina. The muscular coat is characterized by the presence of inner longitudinal and outer circular layers. These layers are indistinct. Amid the muscle cells are seen large, clear, epithelioid-like cells with oval or globular nuclei which are poor in chromatin. The cytoplasm of these cells is either clear or vacuolar and shows no particular tinctorial affinity. It is poor in myofibrils and gives no reaction for glycogen, fat or mucin (Schumacher<sup>5</sup>). Ramifying among these cells are seen anastomosing cells with elongated nuclei and transparent cytoplasm without myofibrils (Masson<sup>1b</sup>). The outer zone of the arteriovenous anastomosis consists of loose delicate collagenous reticulum, in the meshes of which are seen numerous non-medullated nerves. These fibrils are demonstrable with methods of staining with methylene blue (Grant<sup>7a</sup>) and with Cajal's method of retaining with silver (Alvarez and Costero<sup>10</sup>). The entire arteriovenous anastomosis is surrounded by coarse, lamellated collagenous tissue, in the meshes of which are found collecting veins. The primary collecting veins into which the anastomoses drain are characterized by a thin wall almost devoid of muscular cells. These veins surround arteriovenous anastomoses in the form of plexuses. Primary collecting veins open into subpapillary veins and, through the latter, into deeper veins. Quantitative studies of arteriovenous anastomoses give figures

10. Alvarez, C., and Costero, I.: Arch. españ. de oncología 2:391, 1931.

which differ considerably. Grant and Bland's<sup>7b</sup> figures in square centimeters for the index finger are as follows:

Nail bed .....	501
Tip .....	236
Palmar surface of third phalanx.....	150
Palmar surface of second phalanx.....	20
Palmar surface of the first phalanx .....	93
Thenar eminence .....	113
Hypothenar eminence .....	96

Their figures for the foot (second toe) are:

Nail bed .....	593
Pad .....	293
Sole near heel.....	197

As to the size of the individual anastomoses, the following figures in microns for the nail bed are given by Grosser:<sup>4</sup>

Outer transversal region.....	55-65
Inner transversal region.....	18-22

For the finger pulp the figures are:

Outer transversal region.....	90-150
Inner transversal region.....	10-30

The optical section of the arterial portion is narrower than that of the venous portion. From the data reviewed it is evident that arteriovenous anastomoses must be considered as a peculiar peripheral neurovascular anatomic unit of complicated structure and important function.

#### PERSONAL OBSERVATIONS

The serial method employed in my studies not only permitted complete examination and reconstruction of individual arteriovenous anastomoses, but also gave an opportunity to study the entire topographic relation of anastomoses to the surrounding structures. In my description I have adopted the following nomenclature: The portion of the channel connecting artery and vein, or the arteriovenous anastomosis proper, I call the Sucquet-Hoyer canal. The entire anastomotic unit, which includes (1) the afferent artery, (2) the Sucquet-Hoyer canal, (3) the neuroreticular and vascular structures around the Sucquet-Hoyer canal, (4) the outer lamellated collagenous tissue and (5) the primary collecting veins, I call the glomus.

In the first serial sections the periphery of the glomus starts as a round, light area consisting of loose collagenous reticulum and surrounded by coarse, lamellated collagenous tissue. This area is supplied with capillaries, which, for the most part, are cut longitudinally. The

capillaries drain into small slitlike veins, which are located in the meshes of coarse collagenous tissue. Some of these capillaries are twisted around and run along the smaller and larger nerve trunks which are always found at the periphery of the glomus. As soon as the first muscular cells of the Sucquet-Hoyer canal begin to appear, the capillaries become less numerous and less conspicuous. Their endothelial cells intermingle with muscle cells, and only by their large size and deep staining can they be differentiated from the latter. The muscle cells which appear first are elongated, have fusiform nuclei and are circular in arrangement. On reaching the lumen of the Sucquet-Hoyer canal and in the sections cut transversely, the inner muscle cells are arranged more or less longitudinally, while the outer muscle cells are arranged in circular fashion. No true separate longitudinal and circular muscle layers are found in the walls of the Sucquet-Hoyer canal. Depending on orientation, the state of contraction or relaxation and the level of sectioning, the nuclei of individual muscle cells may appear different in size and shape. These factors also determine the appearance of the cytoplasm in the individual muscle cells. Most of the minute tubular and ringlike structures which are seen in the walls of the Sucquet-Hoyer canal are cytoplasmic extensions of muscle cells cut longitudinally and transversely. Amid the ordinary muscle cells are found large epithelioid cells with clear cytoplasm and round or globular nuclei. Followed in serial sections, these epithelioid-like cells appear to be transversely cut muscle cells with voluminous cytoplasm and slightly elongated polyhedral nuclei.

The entire muscular coat of the Sucquet-Hoyer canal is surrounded with a wide clear zone consisting of delicate collagenous reticulum, and containing numerous nonmedullated nerves. With fresh and well fixed material, the trichrome staining of Masson is most helpful for studies of this neuroreticular clear zone and of the smaller and larger nerve trunks always seen in the outer glomic zone. Because it is uniformly constant and selective in its tinctorial affinity, trichrome staining permits detailed topographic studies in serial sections, and the latter compensates for failure of this staining to disclose nerve endings and neurofibrils.

The silver impregnation methods of Rogers, Foot and Schultze-Stohr, when applied to the cutaneous digital glomus, failed to give constant results in my studies. The Schultze-Stohr silver method, however, gave more reliable and selective impregnation. With the silver impregnation a rich network of neurofibrils in close proximity to the muscular walls of the Sucquet-Hoyer canal is seen. No particular or structurally specific nerve endings are found. On the whole, the results with silver impregnation of the skin of the digits are too inconstant to permit proper deductions. Because of their netlike, interlaced arrangement, unmyelinated nerve fibers of the clear periglomic zone appear in

trichrome-stained sections as homogeneous, structureless tubules and rings from 0.5 to 1.5 microns in thickness. These structures are more numerous around the proximal and middle parts of the Sucquet-Hoyer canal, but are inconspicuous in the distal portions. Almost invariably the neurotubular network, if followed in serial sections, is seen to be connected with very small nerve trunks, which in turn can be followed to the point of their connection with large periglomic nerve trunks. Along the course of these minute tubules and rings are seen small, deeply stained, round and rodlike nuclei of Schwann's sheaths. These nuclei become more numerous on approaching the larger nerve trunks.

The intermuscular reticulum is almost invisible. No true internal or external elastic tunica is found. Near the afferent portion of the Sucquet-Hoyer canal the muscular wall becomes narrower and more compact. At the point where the Sucquet-Hoyer canal begins there is a distinct collagenous ruffle containing an elastic lamina (fig. 1) under the endothelium. This structure must be of physiologic significance, and it is at this point that certain pathologic changes of the Sucquet-Hoyer canal are first observed. The lumen of the main afferent artery, if cut transversely and just behind this point, is irregular and corrugated and shows cushion-like elevations consisting of an elevated intima with a bud of longitudinal muscles underneath. The region of the afferent artery where these endotheliomuscular elevations are seen corresponds topographically to the place of the beginning of from two to five small arterioles. These arterioles always begin proximally to the Sucquet-Hoyer canal. Their capillaries supply the walls of the Sucquet-Hoyer canal, the clear neuroreticular zone around this canal and the nerve trunks of the periglomic zone. In other words, they supply all the constituents of the glomus. I call these arterioles preglomic arterioles. Thus, the cushion-like endotheliomuscular elevations in the afferent artery correspond to the place where the Sucquet-Hoyer canal and the preglomic arterioles begin. Their function is to direct the flow of blood into the Sucquet-Hoyer canal and preglomic arterioles. The proximal parts of preglomic arterioles often travel for some distance in the wall of the Sucquet-Hoyer canal. In such cases preglomic intramural arterioles differ from the Sucquet-Hoyer canal with respect to their narrow round lumen, the presence of an internal elastic lamina and their narrow compact layer of circularly arranged muscle cells. Preglomic arterioles are sometimes situated in the vicinity of the Sucquet-Hoyer canal and may be taken for part of this canal, if examined in a single section. Twistings and angulations of the Sucquet-Hoyer canal create the impression that it divides or breaks into several branches. From serial sections it is evident that the passage of this canal is usually single and that preglomic arterioles at no place communicate with its lumen. Sometimes, however, division of the efferent part of the canal into two or three branches is observed.

## EXPLANATION OF PLATE I

Fig. 1.—Section of the glomus at the point where Sucquet-Hoyer's canal begins. An afferent artery is shown below; in the center is the glomus, with the proximal part of the Sucquet-Hoyer canal furnished with elastic lamina, and in the upper part is a section of the primary collecting vein, with its outer wall richly supplied with elastic fibrils. Weigert's elastin stain; camera lucida drawing; ocul., 2  $\times$ ; obj., 9  $\times$ .

Fig. 2.—The efferent part of the Sucquet-Hoyer canal, with part of the primary collecting vein showing only a few muscular cells and with a clear neuroreticular zone present only around the canal. Masson's trichrome stain; camera lucida drawing; ocul., 2  $\times$ ; obj., 9  $\times$ .

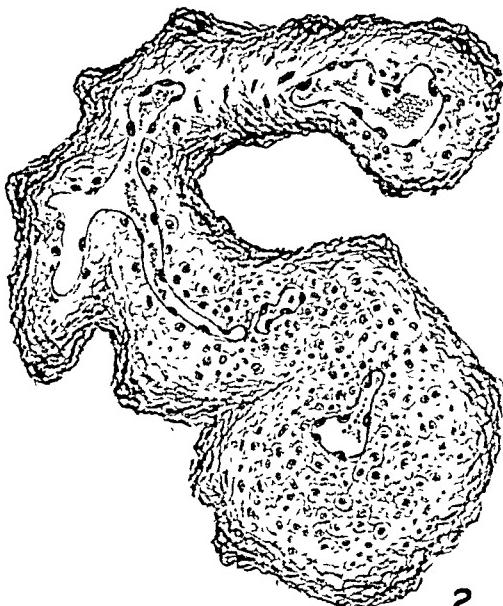
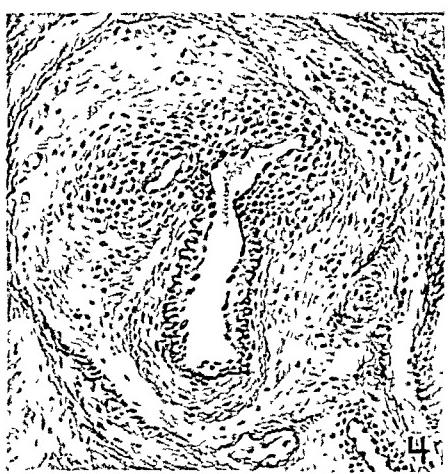
Fig. 3.—The glomus in diabetic gangrene, with hyalinization of the Sucquet-Hoyer canal, which is cut at various levels. The preglomeric arterioles, seen on the left side of the glomus, are markedly thickened, hyalinized and dilated. The periglomeric, clear neuroreticular zone is replaced with thick bundles of collagenous tissue. The primary collecting vein appears as a long, wide cleft encircling the glomus from the right side. The secondary collecting vein is seen in the lower right corner; it is cut transversely and is widely dilated. Masson's trichrome staining; camera lucida drawing; ocul., 10  $\times$ ; obj., 16  $\times$ .

Fig. 4.—The glomus in arteriosclerotic gangrene, with hyalinization of the afferent artery, thickening, distortion and breaking of the elastic fibrils and consequent collapse and permanent dilatation of the Sucquet-Hoyer canal. The periglomeric area shows massive sclerosis. Camera lucida drawing; ocul., 10  $\times$ ; obj., 16  $\times$ . Figure 18 shows same field under lower magnification.

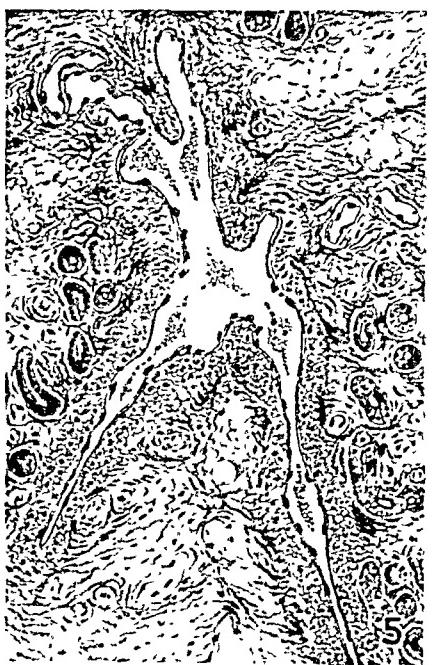
Fig. 5.—Camera lucida drawing made from section 13 of figure 23 and illustrating the relation of digital arteriovenous anastomoses in thrombo-angiitis obliterans. It shows: a small valvular vein ( $V1$ ) in the upper right corner; second vein ( $V2$ ) in the upper left corner ( $V2$  shows a distinctly visible valve at its top); another vein ( $V3$ ) at the bottom; the anastomosing artery  $A1$  approaching the vein ( $V3$ ) formed by the union of the two veins mentioned ( $V1$  and  $V2$ ) from below on the right side and a second anastomosing artery ( $A2$ ) approaching the vein  $V3$  from below on the left side. The anastomosing arteries  $A1$  and  $A2$  show fusiform dilatation. Masson's trichrome stain; ocul., 5  $\times$ ; obj., 16  $\times$ .



1



2



5



Photomicrographs 6 to 13 were taken from serial sections. The corresponding numbers of these serial sections are 45, 44, 43, 42, 40, 38, 35 and 30. Figure 6 shows the place of the cushion-like endotheliomuscular elevations in the afferent artery, which is seen in the center of the field. The arterial lumen is single, but irregular and corrugated. Figures 7 and 8 demonstrate the formation of two separate lumens—one (on the right) of a Sucquet-Hoyer canal and the other (on the left) of a preglomic arteriole. Figures 9, 10 and 11 show the small round lumen of a preglomic arteriole in the upper left quadrant. The preglomic arteriole runs independently in the wall of the Sucquet-Hoyer canal, and the canal follows its own way. In figure 12 the muscle cells of the Sucquet-Hoyer canal begin to disappear, while the lumen of a preglomic arteriole is still seen in the same quadrant. In figure 13 the muscle cells of the Sucquet-Hoyer canal disappear entirely, and the preglomic arteriole divides into several capillaries, which supply the entire glomic unit and the periglomic nerve trunks. (One of the nerve trunks with a capillary is seen in the lower quadrant of the glomus a little to the left.) Figure 14 demonstrates a coiled Sucquet-Hoyer canal cut at different levels. The afferent artery is seen in slightly oblique sectioning. The efferent part of the canal is shown in the upper left corner as an irregular opening with a thin wall. One of the preglomic arterioles is cut longitudinally and is directed toward the periglomic nerve trunk. Several capillary openings of preglomic arterioles are seen in the periglomic zone.

The efferent, or venous, part of the Sucquet-Hoyer canal is characterized by a thin wall with a few muscular cells, by disappearance of a clear neuroreticular zone and by an abundance of elastic tissue in its outer coat (fig. 2). The efferent part of the canal and the capillaries of preglomic arterioles enter the same collecting vein. The width of the primary collecting vein, when empty, is almost that of an ordinary capillary (from 8 to 15 microns). If cut longitudinally, it appears like a capillary slit from 200 to 300 microns in length. It can be differentiated from the capillary only by examination in serial sections. The vascular slit of a longitudinally cut vein can be followed in 15 consecutive sections 7 microns in thickness. Only 1 or 2 sections 7 microns thick are necessary to cover the longitudinally cut capillary. On reconstruction from the serial sections, it was seen that the collecting vein has the shape of a broad ruffle or cape, which encircles the glomus and forms a voluminous receptaculum with a highly developed surface area. In figure 7 such orientation of the primary collecting vein is shown conspicuously, and the vein appears as a long perpendicular slit on the left side of the glomus. This venous slit appears in figures 6 to 12, but is shown best in figure 7. Primary collecting veins are richly supplied with elastic tissue, but they are poor in muscle cells. These veins open

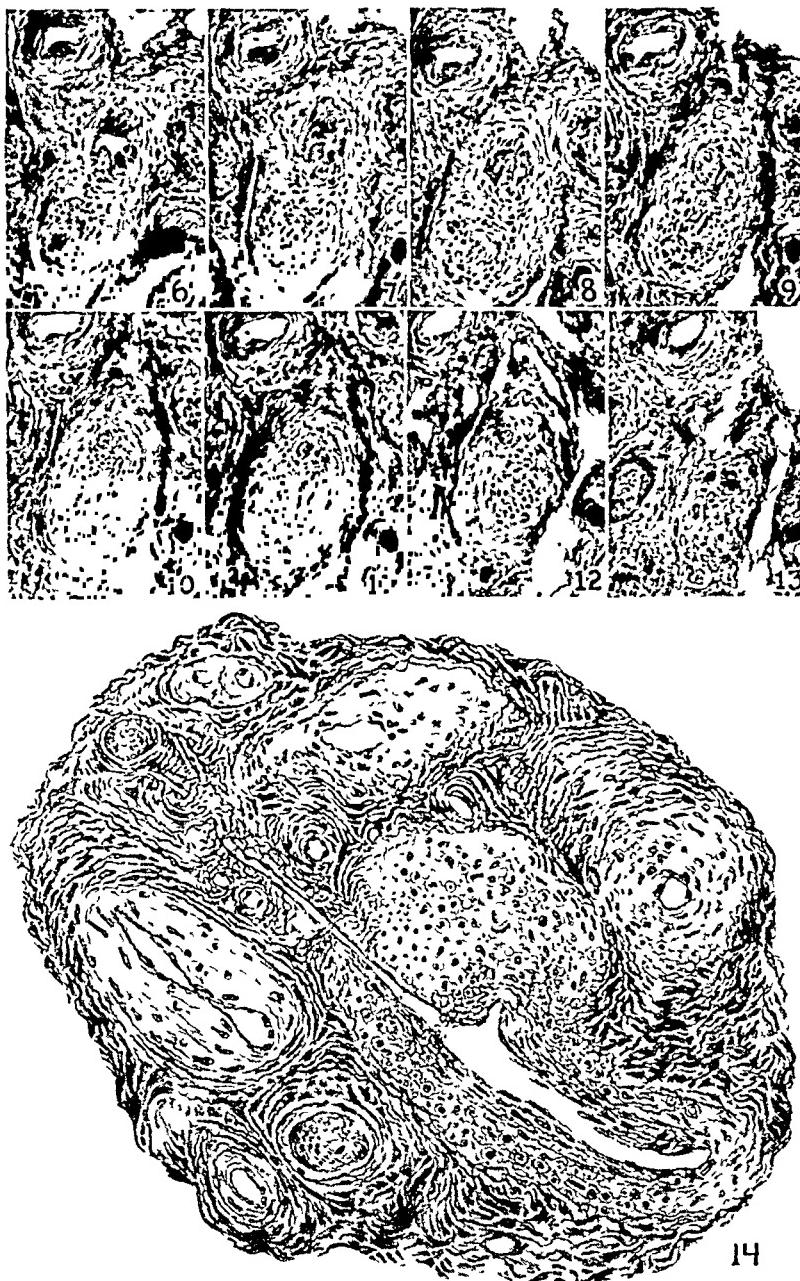
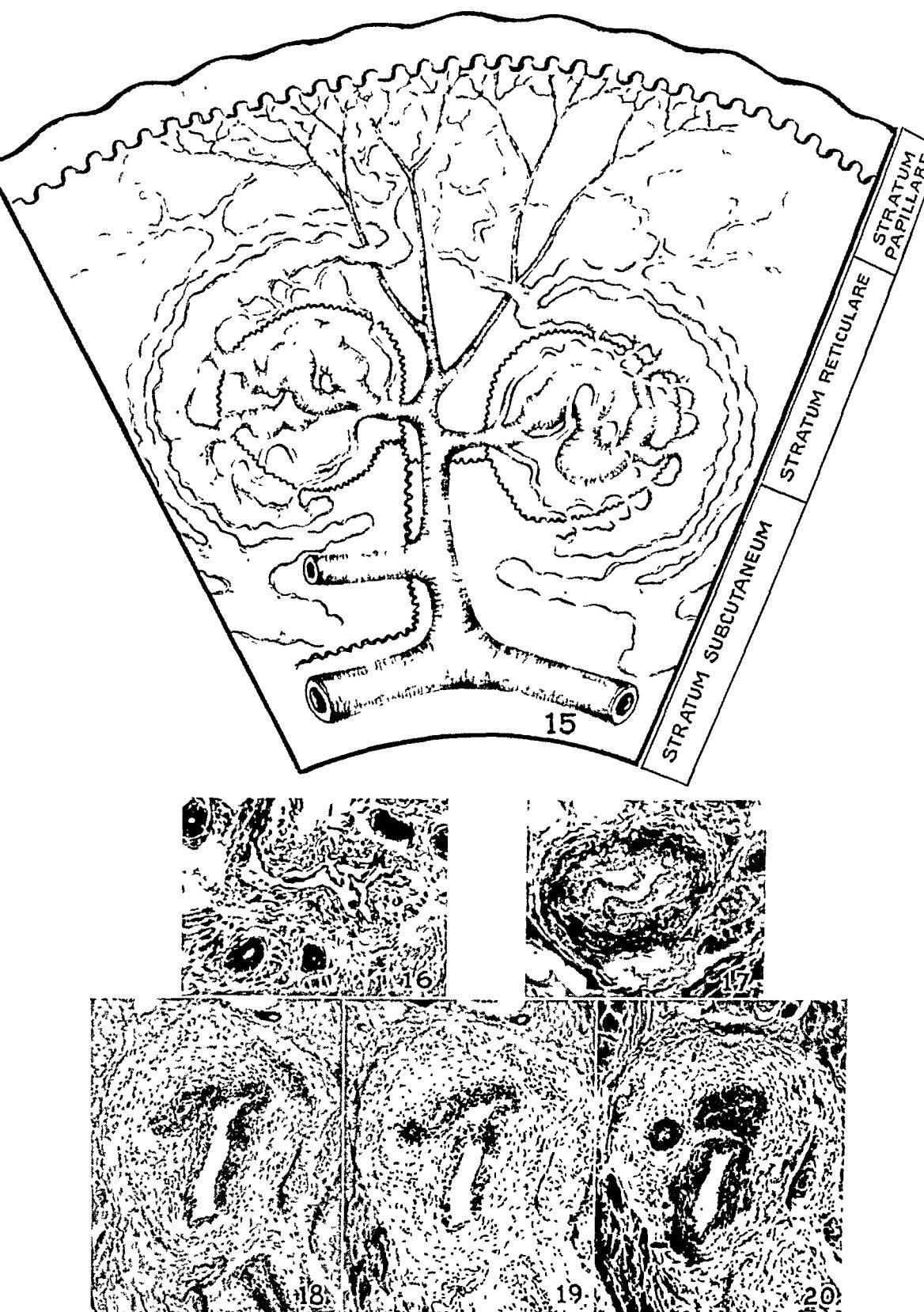


Plate 2

## EXPLANATION OF PLATE 2

Figs. 6 to 13.—Photomicrographs from corresponding serial sections 45, 44, 43, 42, 40, 38, 35 and 30. Figure 6 shows the afferent artery (in the center), its lumen being irregular on account of endotheliomuscular elevations. Figures 7 and 8 show separation of two lumens, the one on the right of the Sucquet-Hoyer canal and the one on the left of the preglomeric arteriole. In figures 9, 10 and 11 an intramural preglomeric arteriole is seen in the upper left quadrant, while the Sucquet-Hoyer canal follows its own way. Figure 12 shows the periphery of the glomus; only a few muscular cells are seen. In figure 13 the muscle cells of the Sucquet-Hoyer canal have disappeared entirely, and the preglomeric arteriole breaks into capillaries supplying all the constituents of the glomus, including preglomeric nerve trunks. One of the nerve trunks is seen in the lower pole of the glomus, and the capillary approaching it is shown. The primary collecting vein is located on the left side of the glomus. It is best seen in figure 7 and appears as a perpendicular slit encircling the glomus from the left side. This venous slit is seen in consecutive serial sections 45 to 35. Masson's trichrome stain; ocul., 10 X; obj., 16 X.

Fig. 14.—A coiled Sucquet-Hoyer canal cut at different levels. The afferent artery is below on the right in slightly oblique sectioning. The proximal part of the canal is shown in the center, with a clear periglomeric neuroreticular zone. The distal opening of the efferent part of the canal is in the upper middle quadrant. One of the preglomeric arterioles, cut longitudinally, is directed toward the nerve trunk, shown in the upper left quadrant. Several small capillary openings of the preglomeric arterioles, supplying all the constituents of the glomus, are shown. Masson's trichrome stain; camera lucida drawing; ocul., 2 X; obj., 9 X.



## EXPLANATION OF PLATE 3

Fig. 15.—Diagrammatic presentation of vascular arrangement and the glomus, as found in the ventral surface of the digit. It shows: (1) all the zones of the skin, including that occupied by the glomeric apparatus; (2) the afferent artery of the glomus; (3) the coiled type of Sucquet-Hoyer canal, characterized by a thick wall; (4) the efferent part of the Sucquet-Hoyer canal entering the primary collecting vein, with the latter appearing as a long, wide ruffle encircling the glomus; (5) the relation of the primary collecting vein to other veins; (6) the system of preglomeric arterioles supplying all the constituents of the glomus and emptying into the primary collecting vein, and (7) division of the periarterial nerve trunks, with branches going to the glomus. This diagram serves to explain arteriovenous and trophic disturbances caused by functional disability and organic destruction of either the entire glomus or one of its constituents.

Fig. 16.—Photomicrograph showing vein of amuscular type of the stratum reticulare, measuring 60 by 120 microns, with 2 valves of the ostial type. Ocul., 10  $\times$ ; obj., 16  $\times$ .

Fig. 17.—Muscular type of vein of the stratum subcutaneum, measuring 240 by 400 microns, with two parietal valves supplied with elastic fibrils. Masson's trichrome stain controlled for elastin; ocul., 7.5  $\times$ ; obj., 16  $\times$ .

Figs. 18, 19 and 20.—Photomicrographs of corresponding serial sections 22, 21 and 16. These three figures demonstrate the proximal part of the digital glomus as found in senile arteriosclerosis. The afferent glomeric artery is sclerosed and shows none of the endotheliomuscular elevations normally seen, which are demonstrated in figure 6. The proximal part of the Sucquet-Hoyer canal is wide open (fig. 18). The periglomeric, clear neuroreticular zone is replaced with dense collagenous tissue. The periglomeric nerve trunks seen on the left, and especially on the right, side of the glomus, show massive lamellated fibrosis. Figures 18 and 19 show sections stained with Weigert's method for elastin. Figure 20 shows a section stained with Masson's trichrome method. Ocul., 7.5  $\times$ ; obj., 16  $\times$ . Figure 18 should be compared with figure 4, which shows the same field under higher magnification.

into subpapillary veins and thence into deep veins. The collecting veins of the stratum reticulare and stratum subcutaneum are furnished with valves. The statement made in a standard book of anatomy that the smaller veins, i. e., those less than 2 mm. in diameter, have no valves is incorrect. Even very small peripheral digital veins show the presence of valves. Figure 16 shows a vein in the inner zone of the stratum reticulare, with two distinctly visible valves of the ostial type. This particular vein measures 60 by 120 microns. Its wall is thin and practically devoid of muscle cells. The valves of the peripheral digital veins are mainly of the ostial type. In smaller veins they consist of delicate connective tissue membrane lined on both sides with endothelial cells. The valves of the large veins have, in addition, fine subendothelial elastic fibrils. Elastic fibrils are better differentiated on the inner surface of the valves than on the outer surface facing the valvular sinus. Figure 17 shows a larger vein of the stratum subcutaneum with a valve of the parietal type, the latter being supplied with elastic fibrils.

Figure 15 recapitulates diagrammatically the anatomic observations described. It shows all zones of the skin, including the zone occupied by glomeric formations. The entire arteriovenous system of the ventral surface of the digit is presented topographically. The relation of the Sucquet-Hoyer canal to the afferent artery and the preglomeric arterioles is indicated. The primary collecting vein of the glomus and its relation to the peripheral and deeper veins are shown. The distribution of the periglomeric nerves and their relation to the capillaries of the preglomeric arterioles are seen.

My studies on the quantitative distribution of the Sucquet-Hoyer canal covered the terminal (ungual) phalanx of the hand and foot. Countings were made from the following regions: the ventral surface, both lateral surfaces, the nail bed and the nail matrix. The digital glomus presents an unstable anatomic unit, and the quantitative results are influenced by a number of factors, of which the age of the person and the pathologic processes are the most important. Examination of the digital vascular system in fetuses from the age of 4½ months to term disclosed complete absence of the Sucquet-Hoyer canal. While sweat glands, nerve trunks and tactile bodies appeared well developed, no formation similar to the Sucquet-Hoyer canal of normal adults was found. In persons over 60 years of age the number of digital Sucquet-Hoyer canals per square centimeter begins to decrease, and this decrease progresses with advance in age. An estimate of the average number of these canals per square centimeter of surface area in the big toe of a normal person 20 years old gives the following figures: ventral surface, 18; lateral surface, 10; nail bed, 24, and nail matrix, 12. The figures obtained from corresponding parts of the thumb are approximately the same. These figures are strikingly lower

than those of Grant and Bland<sup>7b</sup> which have already been cited. My calculations are based on the examination of each individual glomus, not as it appears in a single section, but as it is followed from the beginning to the end in serial sections. I took for arteriovenous anastomoses or for true Sucquet-Hoyer anastomoses only the anatomic structures which were formed in accordance with the descriptions of Sucquet, Hoyer and Grosser. Unmolested by intravascular injections under pressure of washing, fixing and coloring fluids, my fresh surgical material, fixed *in situ*, revealed the state of the vascular digital system as it existed at the time of fixation. Though laborious, my method gives an accurate idea as to the appearance of the glomus at different levels of sectioning. With this method the chance of recounting Sucquet-Hoyer canals is removed, and mistaking anastomoses for short preglomeric arterioles, which run in close proximity to the canals and drain into the same collecting vein, is avoided. The figures of Grant and Bland<sup>7b</sup> were obtained with the method of intravascular injection of washing, fixing and coloring fluids. Their countings were made on cadavers within twenty-four hours after death.

In measuring individual Sucquet-Hoyer canals, I considered a clear periglomeric zone as an integral part of a normal glomus. My figures are: pad of the toe, from 120 to 220 microns, and nail bed, from 60 to 150 microns. The distance from the inner surface of the epidermis to the outer portion of the glomus in digits of average size and development measured from 0.5 to 1 mm. No particular relation of the glomus to the pacinian corpuscles was observed.

From anatomic studies it appears that the digital glomus is a unit of specific structure. The essential characteristics of the Sucquet-Hoyer canal and the entire digital glomus may be summarized as follows:

1. The glomeric system occupies a definite zone in the cutis.
2. One afferent glomeric artery forms from one to four separate Sucquet-Hoyer canals.
3. The beginning of the Sucquet-Hoyer canal is marked by the presence in the afferent artery of cushion-like endotheliomuscular elevations, the function of which is to direct the blood flow into the Sucquet-Hoyer canals and preglomeric arterioles.
4. The wall of the Sucquet-Hoyer canal is rather thick and has a specifically arranged structure.
5. The Sucquet-Hoyer canal is free from elastic tissue, with the exception of its proximal part.
6. The Sucquet-Hoyer canal is surrounded with a clear, wide zone consisting of loose, fine collagenous reticulum enclosing a rich network of nonmedullated nerve fibrils. This clear zone may be considered as

an expansion zone which is furnished with a neuroreticular mechanism controlling the function of the Sucquet-Hoyer canal.

7. The glomic unit is supplied with a specifically arranged system of preglomic arterioles, which nourish all the constituents of the glomus and form an integral part of a functional glomic unit.

8. Primary collecting veins, though lacking in musculature, are richly supplied with elastic tissue. They collect blood not only from the Sucquet-Hoyer canal but also from the preglomic arterioles. Being long and wide, they encircle the glomic unit and thus form a voluminous receptaculum, which has a highly developed surface area.

9. The entire glomus is surrounded with coarse, lamellated collagenous tissue, which separates the glomus from other structures of the cutis.

For proper understanding of the physiology and pathology of the digital glomus, each integral part of it must be taken into consideration.

#### THE GLOMUS IN PATHOLOGIC CONDITIONS

*Inflammation.*—Digits with nonspecific cellulitis from otherwise normal persons were examined. In the early stage of inflammation there are seen in the cutis numerous large, well circumscribed foci of lymphoid infiltration. These foci correspond topographically to the seat of the glomus. They consist of engorged capillaries of preglomic arterioles filled with numerous lymphocytes and polymorphonuclear leukocytes. Massive emigration of these cells through the walls of these capillaries is seen. Following this, both the clear periglomic zone and the muscular wall of the Sucquet-Hoyer canal are infiltrated with inflammatory elements. Muscular cells, especially those of epithelioid type, undergo pyknotic change and begin to decrease in number. No phagocytosis of muscular cells and, owing evidently to the absence of elastic tissue, no formation of foreign body giant cells are noted. The muscular cells simply move to the periphery and lodge in the spaces between the bundles of connective tissue. In the stage of advanced inflammation the wall of the Sucquet-Hoyer canal appears denuded, and its lumen is wide open. Capillaries of preglomic arterioles and the primary collecting veins are distended with stagnant blood. Anatomic changes, caused by inflammation, lead to functional collapse of the glomus. If not completely destroyed, glomic units regain their normal appearance. When damage is irreparable, obliteration of the glomus takes place. In case of local obliteration of the Sucquet-Hoyer canal, a new arteriovenous anastomosis anatomically similar to the original one can be formed. A new Sucquet-Hoyer canal develops from one of the preglomic arterioles. The anatomic factors which favor the development of new canals from preglomic arterioles are: (1) Preglomic arterioles are located in proximity to the clear neuroreticular zone; (2) they are in intimate contact with both the smaller and the larger periglomic nerve trunks, and (3) they are short and open into the same vein which collects the blood from the Sucquet-Hoyer canal. This collecting vein presents a voluminous receptaculum of particular structure and differs in its topographic arrangement from other peripheral veins.

The observations in inflammation indicate that the glomic system is liable to qualitative and quantitative changes. Regressive changes do not necessarily lead to destruction of the Sucquet-Hoyer canals involved. Regeneration of these canals and even formation of new canals can take place.

*Senile Arteriosclerotic Gangrene of the Foot.*—Twenty-four patients were examined, the ages ranging from 60 to 81 years. In this condition general atrophy of the skin and cutaneous appendages is apparent. Of all the structures of the skin, the sweat glands are preserved best. No normal glomic units are observed, and the number of Sucquet-Hoyer canals per square centimeter is greatly reduced. While the average number of Sucquet-Hoyer canals in the tip of the toe of a normal person from 20 to 30 years of age is from 18 to 25 per square centimeter, in the person with senile arteriosclerotic gangrene only from 4 to 6 canals are found for 1 sq. cm. of the corresponding region. Pathologic changes of the Sucquet-Hoyer canals are due mainly to sclerotic thickening of the afferent arteries. The lumens of these arteries show no more cushion-like endotheliomuscular elevations, such as are normally seen at the place where the canals and preglomeric arteries begin. Subendothelial hyalinization, distortion of the elastic lamina and increase in intermuscular collagenous tissue are prominent changes. It is these changes in the afferent artery that initiate the functional disability of the proximal part of the Sucquet-Hoyer canal. Later, preglomeric arterioles undergo hyalinization, and as a result of this the clear neuroreticular periglomic zone is replaced with coarse collagenous tissue. Nerve trunks of the periglomic zone appear surrounded with lamellated, dense fibrous tissue.

Figures 18, 19 and 20, with corresponding serial numbers 22, 21 and 16, demonstrate changes of the Sucquet-Hoyer canal in senile arteriosclerosis, as described. If examined alone and in material poorly fixed and stained with hematoxylin eosin, figure 19 may create the impression that the lumen of the artery is divided into different compartments by bundles of connective tissue. Figure 20 of this series, however, indicates that figure 19 represents the proximal part of the Sucquet-Hoyer canal, which is altered on account of sclerosis of the afferent artery and fibrosis of the periglomic zone. In advanced cases the proximal portion of the Sucquet-Hoyer canal appears sclerotic; the elastic fibers are thickened, broken and distorted, and the entire canal is transformed into a functionally uncontrollable, wide open channel (figs. 4 and 18) through which arterial blood pours continuously into collecting veins. This, in turn, leads to upset of venous drainage in the entire region corresponding to the Sucquet-Hoyer canal involved. In efforts to adjust themselves to the altered mechanical conditions, all the veins of the corresponding region undergo muscular hypertrophy, with secondary dilatation. If sclerosis of the periglomic zone is predominant, the Sucquet-Hoyer canal undergoes obliteration and is replaced with hyalinized fibrous tissue. The valves of the hypertrophied deeper veins are thickened and hyalinized, the elastic fibrils being distorted (fig. 22). These valvular changes add to the disturbances of the venous drainage in the area impaired. The epidermal lining over the region involved shows marked hyperkeratosis. Chronic venous stasis leads to fibrosis and atrophy of the cutaneous appendages. Poorly nourished and diseased areas are predisposed to secondary inflammation, which contributes to more rapid deterioration of the glomic system.

*Diabetic Gangrene of the Foot.*—Nineteen patients with diabetic gangrene were examined. The majority of the cases were complicated by either secondary inflammation or arteriosclerosis, and for these reasons the changes of the glomus demonstrated all possible variants.

Examination of digits free from secondary inflammation and arteriosclerosis showed that in diabetic gangrene the glomus undergoes rather specific changes. In early cases, in which the afferent arteries are not much altered and in which primary collecting veins still hold their size and shape, the primary change is observed in the proximal part of the Sucquet-Hoyer canal, at the place where normally (fig. 1) a

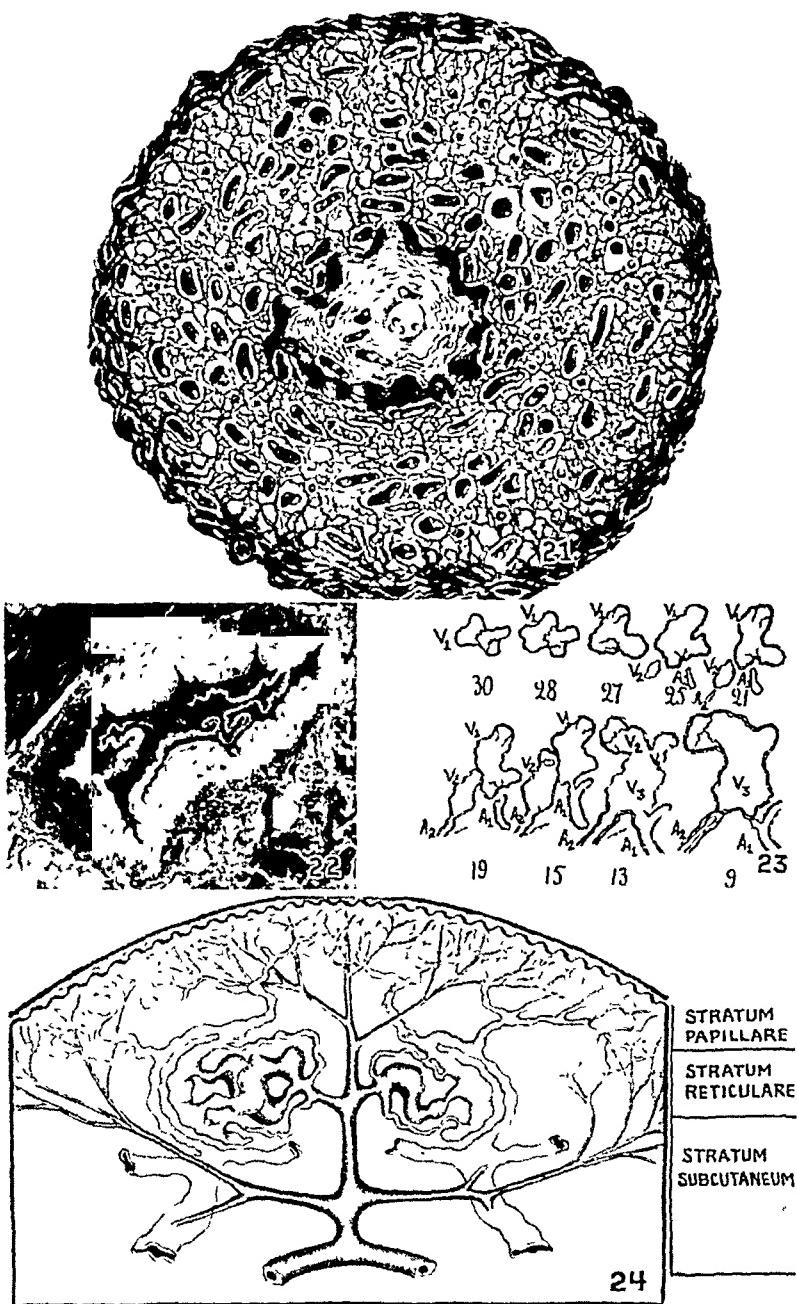


Plate 4

## EXPLANATION OF PLATE 4

Fig. 21.—Proximal part of the Sucquet-Hoyer canal in diabetic gangrene. The subendothelial collagenous ring, which is normal in the center of figure 1, appears thickened and hyalinized; the elastic fibrils are broken; the intermuscular collagenic tissue is increased; the periglomic neuroreticular zone is invaded with sprouts of collagenous tissue, and the outer periglomic zone shows massive lamellated fibrosis. Masson's trichrome stain; camera lucida drawing; ocul., 2 X; obj., 2 X.

Fig. 22.—Valvular insufficiency in a dilated and hypertrophied vein of the stratum subcutaneum from a patient with senile arteriosclerosis. The valves appear thickened and hyalinized, and their elastic fibrils are distorted. Masson's trichrome stain; ocul., 5 X; obj., 16 X.

Fig. 23.—This diagram presents the relation of digital arteriovenous anastomoses found in thrombo-angiitis obliterans and shown by serial sections. The section numbers of the series corresponding to these nine figures are 30, 28, 27, 25, 21, 19, 15, 13 and 9. Section 30 shows a small valvular vein ( $V_1$ ) of the stratum reticulare. Section 25 shows below the vein  $V_1$  a venous space ( $V_2$ ) to the left and an arterial slit ( $A_1$ ) to the right. In section 15 the vein  $V_2$  approaches the vein  $V_1$ . In section 13 the veins  $V_1$  and  $V_2$  unite and form another vein ( $V_3$ ). At the lower left pole of the vein  $V_2$  is seen an artery ( $A_2$ ), which begins in section 21 and enters the vein  $V_2$  in section 15. The artery  $A_1$ , which is first shown in section 25, approaches the vein  $V_1$ , bends to the right and in section 13 forms a short, wide lateral communication with the vein  $V_3$ . In section 9 and the following sections the artery  $A_1$  continues as an ordinary artery terminated with a capillary network.

Fig. 24.—The drawing demonstrates two types of anomalous arteriovenous anastomoses found in thrombo-angiitis obliterans. One, to the right and below, presents the lateral type of anastomosis, in which the artery continues and breaks finally into ordinary capillaries. In the other (the terminal type), which is seen below on the left, the artery ends by entering the wall of the vein. The relation to the normal digital arteriovenous anastomosis, or digital glomus, is indicated.

collagenous ring furnished with elastic fibrils is found. This subendothelial collagenous ring becomes thickened and hyalinized, and elastic fibrils appear distorted (fig. 21). The primary subendothelial hyalinization extends later along the entire middle portion of the Sucquet-Hoyer canal, and preglomeric arterioles and capillaries become thickened and hyalinized. This is followed by an increase in intermuscular collagenous tissue. The sprouts of collagenic fibrils invade the periglomeric clear zone, and the fine network of nonmedullated nerves of the periglomeric zone disappears, being replaced by coarse collagenous tissue. Both the epineurium and the perineurium of the periglomeric nerve trunks show massive lamellated fibrosis. With the advance of periglomeric sclerosis and subendothelial hyaline degeneration of the Sucquet-Hoyer canal and the periglomeric arterioles, the primary collecting veins encircling the glomus appear rather voluminous and permanently dilated (fig. 3). The state of the collecting veins mirrors the extent of anatomic destruction and functional disability of the Sucquet-Hoyer canal. The functional disability of this canal puts an abnormal strain on the collecting veins and consequently on the corresponding deeper veins. As a result of this, the veins undergo hypertrophy and assume gradually an arterial character.

In diabetic gangrene degenerative changes of the Sucquet-Hoyer canal are more prominent than those observed in arteriosclerotic gangrene. Obliteration of the Sucquet-Hoyer canal is more common. In a great number of cases the number of these canals is decreased markedly, and sometimes not a single normal glomus can be found. In noncomplicated cases the degenerative changes of the Sucquet-Hoyer canal are not accompanied by lipoidosis, cellular infiltration or thrombosis. Valvular changes of the veins do not differ from those observed in arteriosclerosis. Continuous disturbance in venous drainage results in sclerosis of the skin, atrophy of the cutaneous appendages and hyperkeratosis of the epidermal lining. Chronic stasis in superficial veins leads to the formation of capillary varices (bullae) in papillary bodies and later creates conditions favorable for bacterial invasion.

*Thrombo-Angitis Obliterans.*—Five patients with thrombo-angiitis obliterans were examined; all were under 40 years of age. One case was especially instructive, for it enabled me to study all the digits and the leg, these being obtained from two consecutive operations.<sup>11</sup> At the time of the first operation the fourth and fifth toes of the left foot were removed because of ulceration and impending gangrene. Five weeks later, amputation of the leg was performed on account of gangrene of the third toe. In all my cases both the digits affected by gangrene and those now affected were examined. Examination of all the regions was made in accordance with the method already described.

Pathologic changes are distributed unevenly in thrombo-angiitis obliterans, and vary greatly. One pathologic manifestation, which appears to be characteristic of this disease and which is commonly observed in both grossly affected and nonaffected digits, is generalized massive fibrosis. Transformation of fibrillar interstitial connective tissue into swollen collagenous tissue justly permits the use of the term interstitial sclerosis. This sclerosis involves all the zones of the skin. It affects all the appendages of the skin to such an extent that fibrosis and atrophy in very old persons with senile arteriosclerosis may be considered as insignificant.

11. Dr. Walter A. Calihan of the Highland Hospital of Rochester furnished this case, which was supplemented with a complete history, including a report of all the modern diagnostic methods.

The state of the glomic system varies in different regions of the same digit. In the region where thrombosis and secondary inflammation are absent and where other changes of the skin are not prominent the following histologic picture is seen: The Sucquet-Hoyer canals are free from primary degenerative changes; the intimal and muscular layers are not altered. When subjected to pressure by surrounding sclerosed tissue, the glomic units appear compressed and diminished in size. The periglomic neuroreticular zone is reduced greatly; dense fibrous tissue encroaches on the capillaries of the preglomic arterioles and the primary collecting veins, and the epineurium and perineurium of the periglomic nerve trunks show extensive lamellated fibrosis. With the exception of massive periglomic sclerosis and sclerosis of the nerve trunks, no other changes are observed which would contribute to functional disability of the Sucquet-Hoyer canals.

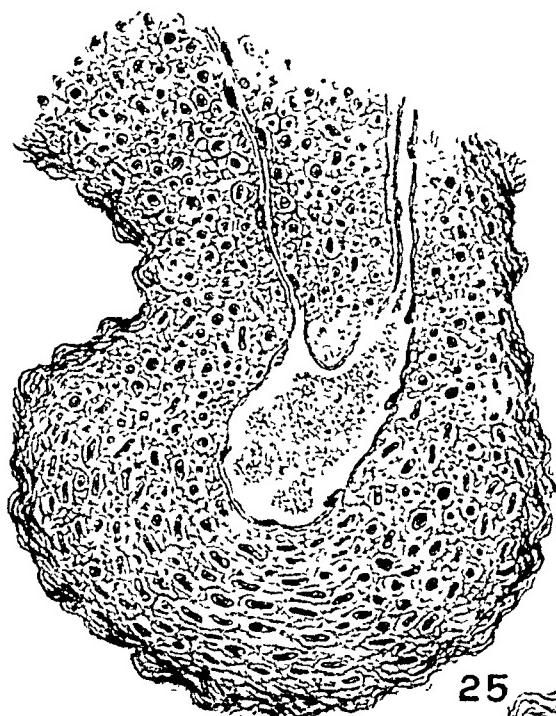
In the regions where thrombosis is prominent the glomic system is affected by thrombosis, both directly and indirectly. The thrombi observed in the peripheral digital system are small and multiple. The average length of the thrombi found in the Sucquet-Hoyer canals varies from 25 to 70 microns. As to the location of thrombi, the following peculiarities are observed:

1. The thrombus may be lodged in the afferent artery, obstructing both the Sucquet-Hoyer canals and the preglomic arterioles.
2. The thrombus may be lodged in the Sucquet-Hoyer canal itself, while preglomic arterioles are free from thrombi.
3. The thrombus may be lodged in the afferent artery, obstructing the intramural preglomic arteriole, but not extending into the Sucquet-Hoyer canal (fig. 25).
4. Of several neighboring Sucquet-Hoyer canals formed by the same afferent artery, one may be blocked by a thrombus and the others may remain free from thrombi.
5. The thrombus may be lodged in a perineural or intraneurial capillary of one of the periglomic nerve trunks.

The described localization of glomic thrombi and the chance that some of them may be septic contribute to a variety of glomic changes, the functional importance of which is obvious.

The thrombi in the larger deeper arteries are of considerable size, but they are also multiple and are lodged at various levels of the same artery. This facilitates efforts to establish collateral circulation through the vasa vasorum. If proper histologic technic is employed, the vasa vasorum of the digital arteries are seen to penetrate to the intima.

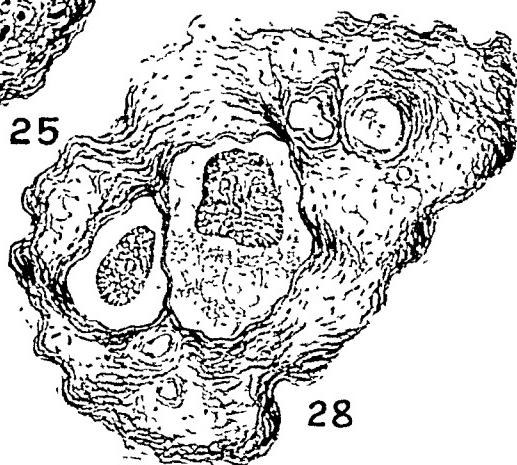
In certain areas the entire arteriovenous system appears altered in a peculiar fashion. The arteries are tortuous and thinned; their intermuscular collagenous tissue is prominent, and, if cut longitudinally, the arteries show regional fusiform dilatation. The veins of both smaller and larger caliber in such regions are widely dilated; their muscular layer is in a hypertrophic state, and elastic tissue is more abundant and is distributed irregularly. Since no local inflammatory or organic obstructive lesions in the glomic system, which could explain the observed vascular changes, were observed in my study, careful examination of sections of a long series was made. On reconstruction of the arteriovenous plexus, it was seen that certain arterial branches of the stratum reticulare and stratum subcutaneum have direct communication with the veins. These communications are entirely different from the Sucquet-Hoyer canals. Two types of arteriovenous communication were seen. In the first type an arterial branch ends by entering the vein through its wall. In the second type an artery which runs parallel to the vein approaches the



25



27



28



26



29

Plate 5

## EXPLANATION OF PLATE 5

These figures illustrate various conditions found in thrombo-angiitis obliterans.

Fig. 25.—The thrombus obstructs the afferent artery and in the center extends to the preglomic arteriole (upper right), while the Sucquet-Hoyer canal (upper left) is closed and free from thrombosis. Masson's trichrome stain; camera lucida drawing; ocul., 10  $\times$ ; obj., 4  $\times$ .

Fig. 26.—Photomicrograph showing an artery of the stratum subcutaneum, with fusiform dilatation and thrombus due to stagnation. Neither inflammatory nor degenerative changes are seen in the artery at the place of fusiform dilatation. Masson's trichrome stain; ocul., 5  $\times$ ; obj., 16  $\times$ .

Fig. 27.—Photomicrograph showing: massive sclerosis of the stratum reticulare; all stages of dilatation of the capillaries, with formation in the papillary bodies of capillary varices lined with prickle cells (hemorrhagic bullae), and extensive hyperkeratosis of the epidermal lining. The entire picture resembles closely angiokeratoma of Mibelli. Masson's trichrome stain; filter H was used to accentuate the capillary varices; ocul., 5  $\times$ ; obj., 16  $\times$ .

Fig. 28.—Results of perineural and intraneurial capillary varices, indicated on the left by distention of the perineural space and an accumulation in it of acellular fluid (hydroneurocele) and in the center by distention of the perineural space, with hemorrhage into it (hematoneurocele). Masson's trichrome stain; camera lucida drawing; ocul., 10  $\times$ ; obj., 9  $\times$ .

Fig. 29.—Photomicrograph showing the valvular vein of the outer zone of the stratum subcutaneum. On the right side, in the lumen, behind the ostial valve is seen an accumulation of inflammatory cells brought by a contributory vein which drains a septic region. Here, behind the valve in the valvular sinus, phlebitis and the formation of a venous thrombus are beginning. Masson's trichrome stain; ocul., 10  $\times$ ; obj., 16  $\times$ .

vein closely and forms a short lateral communication leading into it. The artery continues as an ordinary artery and ends with the formation of a capillary network. In the first, or terminal, type of arteriovenous communication the arterial communicating branch is from 0.5 to 1.5 mm. long and from 100 to 250 microns in diameter. In the second, or lateral, type of arteriovenous communication the lateral passage between the artery and the vein is from 200 to 350 microns long and from 100 to 160 microns in diameter.

Figure 23 presents the relation of arteriovenous communications, as shown by serial sections. Section 30 begins with a small valvular vein (*V1*) situated in the inner zone of the stratum reticulare, which continues in sections 28 and 27. In section 25 there appears in the lower extravenous region a small venous space (*V2*) on the left and a small arterial slit (*A1*) on the right. The lower left venous space (*V2*) becomes larger and in section 15 approaches closely the wall of the vein *V1*. At the lower left pole of the vein *V2* is an artery (*A2*), which begins in section 21 and which is seen entering the vein *V2* in section 15. In section 13 the septum between the two venous spaces disappears, and veins *V1* and *V2* unite and form a large vein (*V3*). The artery *A1*, which is first shown in section 25, on approaching the vein *V1*, bends to the right and in section 13 forms a wide, short lateral communication with an open channel leading into the large vein (*V3*). The artery *A1* then continues as an ordinary artery and terminates with a capillary network.

The orientation of vessels *A1* and *A2*, the direction of their branches, the arrangement of the elastic fibrils and the structure of the muscular layer are all characteristic of the arterial type of these vessels. The appearance in serial sections, the type of distribution of the elastic fibrils, the structure of the muscular coat and of the adventitia and the presence of the valves leave no doubt that the vascular spaces *V1*, *V2* and *V3* are true veins.

Figure 5 is taken from section 13 of the schematically presented series just discussed (fig. 23). The upper right corner of the large vascular space shows the vein *V1*. The upper left corner of this space shows the vein *V2*, with a distinctly visible valve at its top. The lower part of the vascular space corresponds to the vein *V3*. At the bottom of the venous space *V3* are seen the artery *A1* to the right and artery *A2* to the left. A schematic presentation of the two types of peripheral digital arteriovenous communications is shown in figure 24. This figure indicates the topographic relation of the described anomalous arteriovenous anastomosis to the normal arteriovenous anastomosis, or the normal glomus.

Of particular significance is the fusiform dilatation of the artery proximal to the anastomosis (fig. 5). Though this dilatation is most conspicuous in the region nearest the place where the artery enters the vein, other parts of the anastomotic artery may show fusiform dilatation of similar character. The vein *V3*, which is proximal to the place of arteriovenous union, is considerably dilated, and its wall shows an increase in elastic tissue. The artery at the place of fusiform dilatation shows considerable increase in intermuscular collagenous tissue. The elastic fibrils appear thinned and stretched, but no actual increase in elastic tissue is observed in the dilated portion of the artery.

On further examination, the fusiform dilatations were seen not only in the arterial branches directly communicating with the vein, but in the deeper, larger arteries as well. Since no primary inflammatory or degenerative changes were seen at the places of fusiform dilatation of the arteries, it is evident that the process of dilatation must be slow and must be due to some continuous mechanical interference with normal circulation. The only causative agent that I consider as

fully adequate to explain this interference is an anatomic abnormality manifested by the presence of arteriovenous anastomoses. These anomalous anastomoses are entirely different from the arteriovenous anastomoses which were described by Sucquet and Hoyer and which are normally present in the peripheral digital system.

It must be mentioned once again that the changes of the Sucquet-Hoyer canal in the patients with thrombo-angiitis obliterans whom I examined were not primary and were not sufficient to explain the pathologic phenomena observed. Thrombosis of the larger peripheral arteries was of secondary importance. Thrombi may be absent entirely, and yet arterialization of the veins, regional fusiform dilatation of the arteries and other signs of venous stasis may be conspicuous. Sometimes a thrombus is lodged at the place of fusiform dilatation, but neither inflammatory nor degenerative changes of the arterial wall are seen at the point of dilatation (fig. 26). This indicates that such a thrombus is of the type due to stagnation and the lodging of the thrombus in the dilated portion of the artery is due primarily to a mechanical factor causing general slowing of arterial circulation.

Arterialization of the veins is evidently the result of adaptation of the veins to abnormal strain created by an uncontrollable flow of arterial blood through the arteriovenous anastomoses. The existence of anomalous anastomoses explains the mechanism of venous stasis and its sequence, manifested by interstitial sclerosis (sclerodactylia) and atrophy of the cutaneous appendages. The capillaries of the papillary bodies affected by generalized vascular stasis are distended, and large capillary varices are formed in the papillary bodies. If the papillary body contains more than one capillary, the vascular cavity appears divided by septums. As the result of pressure atrophy the connective tissue of papillary bodies disappears, and a vascular cavity lined with prickle cells of the epidermal layer is formed. The horny layer is extremely thickened. At this stage the histologic picture is similar to that of angiokeratoma of Mibelli (fig. 27). Stagnating material of such peripheral capillary varices (bullae) offers a good medium for bacterial growth.

As a result of chronic venous stasis, distention of the perineural and intraneuronal capillaries of both the smaller and the larger nerve trunks takes place. The formation of perineural and intraneuronal capillary varices is followed by local interference with the flow of fluid along the epineural and perineural spaces. The nerve trunks in such regions appear surrounded by a wide space filled with clear acellular fluid. If hemorrhage takes place, the wide perineural space appears filled with blood (fig. 28). Considering the classic work of Key and Retzius<sup>12</sup> and the recent work of Funaoka<sup>13</sup> on the flow of fluid in epineural and perineural spaces in the peripheral nerves, it is permissible to use for the conditions described the terms peripheral hydronurocele and hematoneurocele.

The valves of the veins play a double mechanical rôle. They direct the flow of blood brought by individual smaller veins coming from different regions, and they protect the distal venous column from abuse. In thrombo-angiitis obliterans the valves of dilated and hypertrophied veins appear thickened and hyalinized, with the elastic fibrils distorted. Since head pressure from the anomalous arteriovenous anastomosis continues, the burden of the valves becomes increasingly heavy, and this leads gradually to valvular insufficiency. If one of the contributory veins drains a region affected with inflammation, the larger collecting vein shows an accumulation of inflammatory cells behind the corresponding valve only (fig. 29).

12. Key, A., and Retzius, G.: Studien in der Anatomie des Nervensystems, Arch. f. Micr. Anat. 9:308, 1873.

13. Funaoka, S., quoted by Hassin, G.: Arch. Neurol. & Psychiat. 27:58, 1932.

Phlebitis and incipient formation of a venous thrombus begin in the valvular sinus, i. e., behind the valve which directs the blood coming in from the septic region.

*Supernumerary Digits (Pedunculated Postminimi) in Man.*—Slides from 17 cases of Cummins were examined, the patients ranging from those stillborn at term to those aged 4 days. Only 1 infant was 2 weeks of age. All the details of the histologic observations can be found in the paper by Cummins.<sup>6</sup> My studies were limited mainly to the state of the glomeric system, and in the slides examined I was unable to observe anatomic structures which would correspond to the arteriovenous anastomoses of the Sucquet-Hoyer type.

#### COMMENT

The digital glomus presents a specifically constructed and specifically located system. This system differs from the ordinary circulation, in which arteries and veins are reunited by capillaries. The Sucquet-Hoyer canal is a main part of the glomus. Other constituents of the glomus are: an afferent artery; a periglomeric, clear neuroreticular zone; preglomeric arterioles; periglomeric nerve trunks; an outer, lamellated collagenous zone, and the primary collecting veins. Such an anatomic concept is helpful for proper understanding of the normal function of the Sucquet-Hoyer canal. In pathologic conditions each of the aforementioned constituents of the glomus can be an initiating point in the change of the entire glomeric unit.

The glomus serves two functions—local and general. If the digits are exposed to cold, it is through the agency of the glomeric system that local temperature is maintained and even raised. This is accomplished by diverting blood from the capillaries and rushing it through anastomoses into collecting veins with a highly developed surface area. If, owing to mechanical pressure or other influence, the peripheral arterial pressure becomes abnormally high, the anastomotic by-passages divert the arterial blood and thus relieve the peripheral arterial system of an extra burden. If the peripheral veins and venules are unable to cope with the influx of blood, the anastomoses, by sending a part of the blood directly into the deep veins, help in correcting local disturbances in the venous circulation. Recent investigations by the physiologic school of Lewis<sup>14</sup> prove convincingly that the glomeric system also forms an important factor in the mechanism for regulating the general temperature of the body. When fully opened, it aids the dispersal of heat by allowing an enormous flow of blood to pass through the digits. When closed, it reduces the dispersal of heat to the maximum physiologic limit.

The absence of the glomeric system in a fetus aged from 5 months to term indicates that this system evidently is not needed in fetal life.

14. Lewis, T., and Pickering, G.: Heart 16:33, 1931. Grant,<sup>7a</sup> Grant and Bland.<sup>7b</sup>

Development of the glomic system takes place within the first months of postnatal life. It is known that regulation of heat is one of the least developed functions of premature infants. The body temperature of such infants shows marked fluctuations, with a tendency to hypothermia. This is usually attributed to the following conditions: (1) lack of development of the nervous system and especially of the respiratory center, (2) loss of heat through radiation, (3) weak cardiac action, (4) insufficient combustion of oxygen and (5) improper metabolism. Both anatomic and physiologic knowledge of the glomic system reasonably suggest that the poor control of body temperature in premature infants may be due primarily to absence of the peripheral glomic system. Retarded and improper development of the glomic system in normal infants born at term contributes greatly to the deficiency in controlling the local and general temperature of the body.

In a normal adult each integral part of the glomus is anatomically well differentiated. Each constituent works as a part of one functional unit. Close anatomic and physiologic interrelation of all constituents explains the liability of the glomus to various changes. Regeneration and development of a new Sucquet-Hoyer canal is possible. Generally speaking, formation of new capillaries and subsequent transformation of capillaries into veins and arteries or into an adult vascular pattern is of common occurrence (Clark,<sup>15</sup> Sandison<sup>16</sup>). The process of development of a new and functionally efficient Sucquet-Hoyer canal is much more complicated than an ordinary capillary transformation under experimental conditions. Development of a new canal takes place only in a zone normally occupied by a glomic unit. Preglomic arterioles form a foundation for development of a new canal. These arterioles are part of the afferent glomic artery. They are short and open into specifically orientated veins characterized by a highly developed surface area. They are in close relation to the clear periglomic neuroreticular zone and the periglomic nerve trunks. Such anatomic coordination is most favorable for the development of a new and functionally efficient glomus from preglobular arterioles.

The presented concept on the anatomy of the normal glomus permits one to establish certain differential points in the pathologic anatomy of diseases affecting the digits. In inflammation the digital glomus presents a *locus minoris resistentiac*. Functionally sensitive to thermic, mechanical, chemical and neural influences, the glomus initiates changes leading to venous stasis over the area affected. Stasis in primary collecting veins interferes with the drainage of preglobular arterioles and is followed by inflammatory cellular infiltration into the clear periglomic zone and muscular wall of the Sucquet-Hoyer canal. Loosely arranged,

15. Clark, E.; Hitschler, W. J., and others: Anat. Rec. **50**:129, 1931.

16. Sandison, J.: Am. J. Anat. **41**:475, 1928.

lacking in elastic tissue support and poor in intermuscular collagenous tissue, the muscular cells of the Sucquet-Hoyer canal acquire an embryonal ameboid character and gradually move into periglomic connective tissue. As the result of such regressive remodeling, the Sucquet-Hoyer canal is transformed into a large, distended capillary surrounded with inflammatory elements and containing only a few pyknotic cells. In case of recovery, progressive remodeling, i. e., transformation of the affected Sucquet-Hoyer canal, into the proper glomic pattern takes place. Senile arteriosclerosis affects chiefly the digital arteries of larger caliber. Hyalinization of the afferent glomic arteries is part of a general pathologic process involving all the digital arteries. With the advance of hyaline changes of the afferent glomic artery, the endothelion-muscular elevations, located at the point of origin of the Sucquet-Hoyer canal and the preglomic arterioles, become flattened. This produces functional incapacitation of the proximal part of the Sucquet-Hoyer canal. Extension of hyaline changes to the preglomic arterioles leads to disturbances in nutrition of the entire glomus, including the periglomic nerve trunks. The wall of the Sucquet-Hoyer canal proper retains its structural characteristics for a long time, indicating that this canal is not primarily involved in senile arteriosclerosis. Beginning functional disturbances are due to hyaline changes of the afferent artery, and the anatomic deterioration of the glomus in cases not complicated by inflammation is the result of gradual atrophic changes of all the constituents of the glomus.

Comparative studies on the state of the glomic system in arteriosclerotic and diabetic gangrene do not permit consideration of these two processes as absolutely identical. Current opinion is that the pathologic changes of the vessels in arteriosclerosis and diabetes are the same. Gangrene, according to this opinion, is not due to diabetes *per se* but to a mortifying process dependent on extensive arteriosclerosis. This opinion is based on comparative observations in large arteries and large veins. The majority of the cases of diabetic gangrene are complicated by either secondary inflammation or arteriosclerosis. But if all the digits and all the regions of each affected and nonaffected digit are examined systematically, it becomes difficult to attribute the entire variety of changes observed to sclerosis of the large arteries as a sole cause. If, in cases of diabetes, the digital regions free from gangrene and sclerotic changes in the afferent glomic artery are examined, it is seen that the Sucquet-Hoyer canal and the preglomic arterioles are the first to show signs of primary degenerative change. The intima of the Sucquet-Hoyer canal appears thickened and hyalinized. This degenerative process begins in the proximal part of the canal and extends along the intima of the entire anastomotic passage. In arteriosclerosis the digits free from gangrene and secondary inflammation may not show any appreciable changes in the wall of the

Sucquet-Hoyer canal. Formation of subepithelial hemorrhagic bullae is seen in arteriosclerosis and diabetic gangrene. In both diseases these bullae are due to capillary varices in papillary bodies, caused by functional collapse of the Sucquet-Hoyer canal and consequent interference with the venous drainage over the area affected. Depending on the extent and duration of changes in the Sucquet-Hoyer canal, the corresponding veins undergo dilatation and hypertrophy and become gradually arterialized.

In thrombo-angiitis obliterans no primary degenerative or inflammatory changes of the Sucquet-Hoyer canal are observed. In areas affected by thrombosis the individual glomic units may be the seat of thrombi. The topographic varieties of the glomic thrombi which have been described contribute to the pathologic peculiarities of the glomus affected. It is evident that local changes of the glomus are due either to glomic thrombi or to atrophy caused by generalized interstitial sclerosis of surrounding tissues. Neither generalized interstitial sclerosis nor the vascular changes seen can be explained by observations in the glomic system alone. Nothing is seen that would permit consideration of inflammation of the vessels or of thrombosis as a causative agent. Comparative studies of my entire material show that digital changes in thrombo-angiitis obliterans are specific and must be due to some other cause. On examination of serial sections of all the regions of grossly affected and nonaffected digits, it was seen that the mechanism underlying the entire pathologic picture must be attributed to a vascular anomaly manifested by the presence of arteriovenous anastomoses entirely different from those described by Sucquet and Hoyer. Peripheral venous stasis, dilatation and arterIALIZATION of the veins, thinning and hyalinization of the arteries, fusiform dilatation of the arteries and general atrophic changes appear as a sequence of conditions created by continuous deviation of arterial blood through anastomoses into the veins.

This observation involves the general question of arteriovenous anastomoses. Anatomically, arteriovenous anastomoses are divided into two groups. To the first group belong the arteriovenous anastomoses of Sucquet-Hoyer. These peripheral anastomoses, which are present in normal conditions, are characterized by their coiled, twisted appearance, by their peculiar structure which differs from the structure of arteries, veins and capillaries and by their diameter which never exceeds 0.1 mm. In man these anastomoses are under control of vaso-motor nerves, and their function is to divert rapidly the flow of blood from the artery directly into the veins. For this reason Sucquet chose for these anastomoses the name *canaux dérivatifs*. In case pressure in the arteries and veins becomes high, these anastomoses reestablish normal pressure by opening their communications between the two systems.

In addition, by the same mechanism of opening and closing, they regulate both the local and the general temperature.

The arteriovenous anastomoses of the second group are the direct anastomoses between arteries and veins of larger caliber. In lower animals arteriovenous anastomoses are common. In man arteriovenous anastomoses are always present before birth. Chemical proof of the existence of arteriovenous anastomoses is demonstrated by the high unsaturation of fetal blood in respect to oxygen, which indicates that at term the fetus exists normally in the state of cyanosis (Eastman<sup>17</sup>). In the adult human being arteriovenous anastomoses are rare, and they are considered as a vascular anomaly. All varieties, described both as to type and as to topographic distribution, have been reported in the literature (Tschausoff,<sup>18</sup> Gérard,<sup>19</sup> de Takáts,<sup>20</sup> de Takáts and Mackenzie,<sup>21</sup> Szepsenwol,<sup>22</sup> Miller and Godfrey<sup>23</sup> and others).

Experiments on reproduction of arteriovenous anastomotic anomalies began with the classic work of Carrel and Guthrie.<sup>24</sup> Their experiments demonstrated the remarkable adaptability of arteries and veins to mechanical conditions created by reversing the circulation. Reid<sup>25</sup> has studied systematically the effect of arteriovenous fistulas on the heart and blood vessels. His work includes experimental and clinical investigations, and his observations serve to explain certain observations in my material. He noted that the wall of the vein involved in an arteriovenous fistula becomes dilated and hypertrophied (arterializes) and that an arteriovenous fistula of long duration usually causes dilatation of the artery proximal to the fistula. Callander's<sup>26</sup> publication presents a thorough review of 447 cases of arteriovenous fistulas, with a detailed discussion of their physiology and pathologic anatomy. Of all the theories cited by Callander which are offered as an explanation of arterial dilatation, that of Delbet sounds most reasonable. According to Delbet, the dilatation and thinning of the artery are to be explained on the basis of atrophy of disuse, as the artery in the presence of arteriovenous anastomosis no longer needs to contract against the customary arterial pressure. Among other phenomena associated with arteriovenous fistulas, Callander mentioned a variety of nutritive and

17. Eastman, N.: Bull. Johns Hopkins Hosp. **47**:221, 1930.

18. Tschausoff, M.: Medicinischer Bote no. 15 (in Russian), quoted by Hoyer: Jahrb. f. Anat. u. Physiol. **3**:176, 1874.

19. Gérard, G.: Arch. de physiol. norm. et path. **7**:597, 1895.

20. De Takáts, G.: Surg., Gynec. & Obst. **55**:227, 1932.

21. De Takáts, G., and Mackenzie, W.: Surg., Gynec. & Obst. **58**:655, 1934.

22. Szepsenwol, J.: Arch. d'anat., d'histol. et d'embryol. **15**:45, 1932.

23. Miller, N., and Godfrey, J.: Anat. Rec. **13**:177, 1917.

24. Carrel, A., and Guthrie, C.: Compt. rend. Soc. de biol. **58**:730, 1906.

25. Reid, M.: Bull. Johns Hopkins Hosp. **31**:43, 1920.

26. Callander, C.: Ann. Surg. **71**:428, 1920.

other changes furnished by obstruction of venous return, including edematous infiltration and hypertrophy of the extremities, hypertrophy of subcutaneous tissue followed by subsequent atrophy of the skin, hypertrophy of the hair and nails, a sense of chilliness and cold in the part at or below the region of anastomosis and spontaneous gangrene of the foot and leg. In the light of my observations, all these changes are of special significance.

In pediatric literature the question of trophic changes and spontaneous gangrene are discussed more clinically than pathologically. The pathologic reports are vague and incomplete. Peckham,<sup>27</sup> in a statistical study of 1,461 stillborn fetuses, found that in 28 per cent the cause of death was not demonstrated, and in 19 per cent death was attributed simply to asphyxia. It appears that if the search for vascular anomalies were made more carefully, the diagnosis of asphyxia could be reduced considerably. Fitzwilliams and Vincent<sup>28</sup> reported gangrene of the leg in an 11 day old infant in whom the only change was thickening of the intima of the vascular wall. Thickening of the intima and increase in elastic tissue are characteristic of the first stage of thrombo-angiitis obliterans. According to Krompecher,<sup>29</sup> the increase in elastic and fibrous tissue (*Elastofibrose*) leads to narrowing of the small vessels and is specific for the first stage of thrombo-angiitis obliterans. In view of this, Krompecher called the entire process *Teleangiostenose*. Increase in elastic tissue he attributed to the abnormal activity of vascular elastoblasts. He observed that *Teleangiostenose* affects both the arteries and the veins, and he considered hereditary degeneration of the mesenchymal tissue (*mesenchymale Heredodegeneration*) as a possible etiologic factor. Carrel and Guthrie<sup>30</sup> produced thickening of the intima, increase in elastic tissue and hypertrophy of the muscle cells by diverting arterial blood into the veins. By reversing the circulation, Carrel was able to transform veins into pulsating arteries and arteries into flaccid veins. In the light of these experiments, it is difficult to attribute Krompecher's *Teleangiostenose* to inborn primary overactivity of elastoblasts. Thickening of the intima in thrombo-angiitis obliterans is the result of neither arteriosclerosis occurring with advancing age nor thrombosis with canalization (Brown, Allen and Mahorner<sup>30</sup>).

My studies on the state of the digital vascular system show that certain changes in arteries and veins disclose nothing but functional

27. Peckham, quoted by Eastman.<sup>17</sup>

28. Fitzwilliams and Vincent, quoted by Grulee, C. G.: Bonar, B. E., and Haynes, R.: Clin. pediat. 3:118, 1926.

29. Krompecher, S.: Beitr. z. path. Anat. u. z. allg. Path. 85:647, 1930.

30. Brown, C.; Allen, E., and Mahorner, H.: Thrombo-Angiitis Obliterans, Philadelphia, W. B. Saunders Company, 1928.

adaptation necessitated by altered mechanical conditions. If Sucquet-Hoyer's arteriovenous anastomosis becomes inefficient, it plays the rôle of a functionally uncontrollable arteriovenous fistula, and, depending on the degree of disability and the duration of the uncontrolled activity of the Sucquet-Hoyer canal, the arteriovenous system of the region involved is changed accordingly. The dependent tissues show all signs of trophic disturbance. This happens in inflammation, arteriosclerosis, diabetic gangrene and thrombo-angiitis obliterans. In thrombo-angiitis obliterans I observed, in addition to localized vascular changes caused by collapse of the Sucquet-Hoyer canal, generalized arteriovenous pathologic changes which were unrelated to the Sucquet-Hoyer system of peripheral anastomoses. Thickening of the intima, increase in elastic tissue, fusiform dilatation of the arteries and dilatation and arterialization of the veins are not limited to a zone occupied by Sucquet-Hoyer's anastomoses. The trophic changes are more diffuse and pronounced than those caused by collapse of Sucquet-Hoyer's anastomoses. I attribute these changes to the peripheral arteriovenous anastomoses. These anastomoses are entirely different from those described by Sucquet and Hoyer, and I consider them as a vascular anomaly.

In looking over descriptions of symptomatology in thrombo-angiitis obliterans, it is significant that local chronic cyanosis of the toes may precede gangrene of the foot by months and even years and may be the only objective sign of the disease. Postural color changes in the earliest stages of thrombo-angiitis obliterans occur only in the tips of the toes. Pain while the patient is at rest occurs almost exclusively in the digits and appears before obvious trophic changes can be observed. Coldness of the extremities is described as an annoying and persisting complaint. It is noteworthy also that in 71 of 100 cases of thrombo-angiitis obliterans both lower extremities were involved (Buerger<sup>8</sup>). Since no definite toxic etiologic agent of thrombo-angiitis has been found, infectious and constitutional factors are considered in the literature. Krompecher's<sup>29</sup> concept of *mesenchymale Heredodegeneration* is directed toward a constitutional anomaly. Gruber<sup>31</sup> considered the inborn constitutional peculiarity of the reaction of the blood vessels as a possible etiologic factor. All the peculiarities of the changes in arteries, veins, glomus and capillaries and all the trophic changes of the skin and cutaneous appendages, including the peripheral nerve, in the cases of thrombo-angiitis obliterans studied by me I attribute directly to the presence of the anomalous arteriovenous anastomoses described.

31. Gruber, G.: Ztschr. f. Kreislaufforsch. 23:537, 1931.

In the search for chemical proof of the existence of arteriovenous anastomoses, a comparison of the gaseous content of the blood from the venous arch of the dorsum of the foot, the lower portion of the internal saphenous vein of the foot and the median basilic vein of the arm was made on a patient with thrombo-angiitis obliterans. The blood was taken at normal room temperature, at the same time, with identical technic and while the patient's breathing was shallow, regular and of the usual frequency. The carbon dioxide content, oxygen content, oxygen combining power and oxygen saturation were determined. The readings for the blood from the veins of the affected foot just indicated were: oxygen content, 16.95 per cent, and oxygen saturation, 82.49 per cent. The comparative readings for the blood from the median vein of the arm were: oxygen content, 13.17 per cent, and oxygen saturation, 63.9 per cent. Although the other factors (Goldschmidt and Light<sup>32</sup>) which may influence the gaseous content of the peripheral blood were kept in mind, the comparative readings obtained are of significance. Both the color and the gaseous content of the blood from the veins of the affected foot indicate that the blood examined was mixed arteriovenous blood. The observation of an anomalous peripheral arteriovenous anastomosis offers a reasonable explanation for the changes observed in the venous blood examined.

The difference between normally found peripheral anastomoses of the Sucquet-Hoyer type and the accidental arteriovenous anastomoses of the type of vascular anomaly are summarized in the table.

My studies on the state of the digital glomic system and my observations of the vascular anomaly, manifested by the existence of arteriovenous anastomoses, suggest the need for more careful examination of the vascular system in regard to the anomalies in my material. The search for peripheral arteriovenous anastomoses requires proper fixation of fresh material, trichrome staining controlled by staining for elastin, examination of all the affected and nonaffected digits and careful reconstruction from uninterrupted, long series of sections. The injection method may be tried, but perfectly fresh material should be used, and the observations should be controlled by the method of reconstruction from serial sections. In my normal and pathologic material, covering 840 individual blocks, I observed abnormal digital arteriovenous anastomoses only in cases of thrombo-angiitis obliterans.

The comparative results of my studies on the digital glomus demonstrate that neurovascular and trophic changes observed in the digits in many instances may be due primarily to local changes in the glomus and not to inflammatory, degenerative or obstructive changes in the large arteries and veins of the extremities. It must be kept in mind

32. Goldschmidt, S., and Light, A.: Am. J. Physiol. 72:127, 146 and 173, 1925.

*Comparative Observations in Sucquet-Hoyer's Anastomoses and Anastomoses of the Type of Vascular Anomaly*

	Normal Peripheral Sucquet-Hoyer's Anastomoses of the Extremities	Digital Arteriovenous Anastomoses of the Type of Vascular Anomaly
Distribution	Ventral and lateral parts of the digits, nail bed, nail matrix, thenar and hypothenar eminences of the hand and sole near the heel	The inner zone of the stratum reticulare and the stratum subcutaneum
Appearance	Coiled, twisted, S-like and seldom straight; afferent artery forms from one to four individual anastomoses	Mostly straight; single; of directly terminal or lateral type
Size	The lumen does not exceed 0.1 mm.; average length is from 0.5 to 0.8 mm.	The lumen is larger than 0.1 mm.; the size varies with the caliber of the communicating vessels and the type of anastomosis
Structure	The afferent artery has endothelio-muscular elevations, which direct the flow of the blood into anastomoses and preglomer arterioles; with the exception of its proximal part, the anastomosis has no elastic lamina; the muscular cells of the wall are specific in structure and arrangement; the perianastomotic region is surrounded with a wide clear neuro-reticular zone (expansion zone); the primary collecting veins are shaped like a long broad ruffle; they encircle the glomus and present a voluminous receptaculum with a highly developed surface	The lumen of the proximal part of the anastomosis does not show endothelio-muscular elevations; the structure of the anastomosis is that of the ordinary artery; the structural changes are secondary and are caused by altered mechanical conditions; the structure and shape of the collecting vein are those of an ordinary vein; hypertrophy and dilatation of corresponding collecting veins are secondary
Control	The contraction and dilatation of the anastomosis are under perfect control of the local and general vaso-motor mechanism	Local and general vaso-motor control is only relative and never perfect
Function	Adjusts peripheral arteriovenous system to changes caused by internal and external influences; controls local arterial and venous pressure; regulates local and general temperature	Purposeless and detrimental
Results of Functional Disability	When in a state of functional disability, the opened anastomosis pours the arterial blood continuously into primary collecting veins; the latter become hypertrophied and dilated; subpapillary and corresponding deep veins of the region affected undergo hypertrophy and dilatation, with generalized peripheral venous stasis; all the mesodermal and ectodermal constituents of the region involved show secondary trophic changes; control of local temperature and, to some extent, of general temperature, is deranged	Lacking in normal vaso-motor control and having a larger diameter than a normal anastomosis, the anomalous anastomosis precipitates more profound changes; the gaseous content of the blood becomes abnormal; the arteries show regional fusiform dilatation, which is more conspicuous in the portion of the anastomosis near the vein; the collecting veins undergo arterialization and dilatation; venous stasis is pronounced; peripheral venous stasis leads to the formation of intrapapillary varices (hemorrhagic bullae); the extent of the secondary trophic changes depends on the position and diameter of the anastomosis and the duration of its existence; trophic changes lead gradually to progressive scleroderma, perineural fibrosis and atrophy of the cutaneous appendages and terminate finally with necrobiosis; the temperature in the part at or below the region of the anastomosis is uncontrollable

that changes in the glomic system and the occasional existence of other abnormal arteriovenous anastomoses introduce disturbing elements into the interpretation of capillaroscopic observations of the digits. The local clinical manifestations and diagnostic tests cannot be understood properly without taking into consideration the state of the glomic system and possible anomalies of the peripheral digital vascular system.

#### SUMMARY

The digital glomus is a normal anatomic unit consisting of: (1) an afferent artery; (2) a Sucquet-Hoyer canal, or arteriovenous anastomosis proper; (3) preglomeric arterioles, nourishing all the constituents of the glomus; (4) a clear periglomeric zone, or expansion zone, furnished with a neuroreticular mechanism which controls the function of the Sucquet-Hoyer canal; (5) a specially arranged system of collecting veins, and (6) an outer lamellated collagenous zone surrounding the entire glomus.

The function of the glomus is to control arteriovenous circulation in the digits and to regulate both the local and the general temperature of the body.

The digital glomus is absent during intra-uterine life, and is formed shortly after birth. Both trophic and thermic disturbances, which are common in premature infants and which are found after birth in some infants born at term, may be reasonably attributed to absence or improper development of the glomic system.

No glomus is found in supernumerary digits (pedunculated post-minimi) in man.

In advanced senility the glomic units undergo atrophy, and their number decreases with age.

The glomus is sensitive to various influences and presents a very unstable unit. It may show both regressive and progressive changes. In case of complete collapse and deterioration of the Sucquet-Hoyer canal, a new canal can develop from one of the preglomeric arterioles described.

Comparative studies of the state of the glomus in arteriosclerotic and diabetic gangrene indicate that the changes observed in the glomus in these two conditions affect the glomus in different ways. In arteriosclerotic gangrene the changes in the glomus are due primarily to hyaline degeneration in the afferent artery of the glomus. In diabetic gangrene the intima of the Sucquet-Hoyer canal and that of preglomeric arterioles are the first to show signs of primary degenerative changes, while the intima of the afferent artery of the glomus remains intact.

In the cases of thrombo-angiitis obliterans studied, nothing was observed that would permit consideration of inflammation of the vessels or of thrombosis as a causative agent. The glomic system

appeared to be free from primary specific changes. The changes observed in arteries, veins and capillaries, the trophic changes of the skin and cutaneous appendages, the inflammatory and necrobiotic changes in the digits I attribute directly to a hitherto unknown vascular anomaly of the peripheral digital system. This anomaly is manifested by the presence of abnormal communications, or anastomoses, between peripheral arteries and veins of the digits. Two types of abnormal anastomoses are seen: a lateral and a terminal type. In their size, structure, and topographic distribution, they differ entirely from the normal peripheral digital anastomoses of the Sucquet-Hoyer type.

Consideration of the state of the glomic system and of the anomalous arteriovenous anastomoses described offers a substantial help for an understanding of the vascular and trophic changes observed in the digits.

It is obvious that the action of chemical substances, the capillaroscopic observations, the local clinical manifestations and the results of various diagnostic tests applied to digits cannot be interpreted properly without taking into consideration the anatomic concepts presented.

# MYOCARDIAL CHANGES IN HYPERTENSION

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That hypertension, or abnormally high blood pressure, is linked with hypertrophy of the heart has long been understood. Strangely enough, agreement has not been reached as to what other changes, aside from the hypertrophy, occur in the hypertensive heart. There have been varying opinions even in regard to the mode of enlarging. Pathologists and anatomists first believed that the number of muscle fibers increased. Then the view was favored that the muscle fibers increased in size as well as in number. More recently, Karsner, Saphir and Todd<sup>1</sup> have shown that the muscle fibers increase in size but not in number.

MacKenzie<sup>2</sup> stated that there is an absence of correlation between clinical and pathologic findings in cases of nonvalvular hypertrophy of the heart, the type commonly spoken of as hypertensive hypertrophy. Tawara<sup>3</sup> could find no histologic equivalent of acute cardiac failure. Christian<sup>4</sup> stated that nothing abnormal is found in the myocardium, aside from hypertrophy. Cohn,<sup>5</sup> on the other hand, believed that definite changes, such as fibrosis, occur on a noninflammatory basis.

According to Christian,<sup>4</sup> hypertrophy is an unfavorable clinical sign, pointing to a poor prognosis. He stated that once a heart begins to enlarge, changes start which regularly progress to cardiac decompensation. Fitz Hugh<sup>6</sup> agreed with this idea; he found that hearts weighing over 600 Gm. fail more rapidly than smaller hearts.

Clinically, when a heart without valvular lesions "fails," the condition is called "chronic myocarditis." This is a poor term, however, as Cohn<sup>5</sup> and Bierring<sup>7</sup> pointed out, because there is no inflammatory basis for

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From the Department of Pathology, Cook County Hospital, Dr. R. H. Jaffé, Director.

1. Karsner, H. T.; Saphir, O., and Todd, T. W.: Am. J. Path. **1**:351, 1925.
2. MacKenzie, J.: Diseases of the Heart, New York, Oxford University Press, 1918.
3. Tawara, S.: Das Reizleitungssystem des Säugetierherzens, Jena, Gustav Fischer, 1906.
4. Christian, H. A.: J. A. M. A. **91**:549, 1928.
5. Cohn, A. E.: Am. J. M. Sc. **177**:619, 1929.
6. Fitz Hugh, G.: New England J. Med. **203**:201, 1930.
7. Bierring, W. L.: J. A. M. A. **101**:663, 1933.

the changes. Riesman<sup>8</sup> and Bierring<sup>7</sup> prefer the term myocardiosis, and Brown<sup>9</sup> offered the terms scarring of the myocardium or myocardial fibrosis as preferable.

Fibrosis of the myocardium is a condition which has been studied since Morgagni<sup>10</sup> first reported a case. Ricord<sup>11</sup> and Virchow<sup>11</sup> called attention to the influence of syphilis in its causation. Ziegler,<sup>12</sup> Turner<sup>11</sup> and Martin<sup>11</sup> pointed out the importance of coronary disease. Brown,<sup>9</sup> in a recent study on the pathogenesis of myocardial fibrosis, divided the causes into three general groups: infectious myocarditis, toxic myocarditis and interference with the blood supply.

By the term myocardial fibrosis in this study is meant the definition which Clawson<sup>13</sup> gave of atrophy of muscle fibers and replacement by scar tissue. Most typically the fibrous tissue is arranged in small islands which often enclose single muscle fibers or fragments of fibers in various stages of atrophy and degeneration.

Because of the lack of agreement as to the myocardial changes in hypertension this study was undertaken. Special attention is given to myocardial fibrosis, because I have frequently observed widespread minute macroscopic and microscopic scars in hypertensive hearts.

#### STATISTICS

This study was made on 27 hearts chosen from a group of 327 hearts of adults after postmortem examinations performed at the Cook County Hospital. These 27 are 8.2 per cent of the total, which gives an idea of the incidence of hypertension among that group of adults.

The 27 hearts were chosen on the basis of weight. With 3 exceptions (cases 9, 11 and 15), all weighed 500 Gm. or more. In the 3 exceptions the weight was only slightly under 500 Gm. This weight was considered indicative of hypertensive hypertrophy. Hearts with old rheumatic deformities of the valves were not studied in order to keep clear of any old myocardial inflammatory lesions. The hearts were fixed in toto in a 10 per cent solution of formaldehyde for two days. Then blocks were cut from seven places, namely, the anterior and posterior walls of the left ventricle, the large anterior papillary muscle in the left ventricle, the interventricular septum near the apex, the anterior wall of the right ventricle and both auricles. Sections from these locations were stained with van Gieson's stain and studied for myocardial fibrosis, which was classed from a trace to four plus, when present. The degree of endocardial sclerosis was also noted. The medium-sized and smaller arteries and the arterioles were studied in all the sections. Representative sections were stained with Weigert's elastic tissue stain.

8. Riesman, D.: M. Clin. North America **10**:361, 1926.

9. Brown, M. R.: Am. J. M. Sc. **184**:707, 1932.

10. Morgagni, J. B.: De sedibus et causis morborum, Naples, 1762.

11. Quoted by Cowan.<sup>21</sup>

12. Ziegler, E.: Deutsches Arch. f. klin. Med. **25**:586, 1880.

13. Clawson, B. J.: Am. Heart J. **4**:1, 1928.

## RESULTS

In the table are listed the essential data in each case. The case number, race, sex, age and blood pressure are given in the first five columns. Next in order are the weight of the heart in grams, the thickness of the left ventricle in millimeters, the thickness of the right ventricle in millimeters, any related myocardial or vascular changes and the cause of death.

In the next column the amount of sclerosis of the large coronary arteries is listed, being classed as marked, moderate or slight. The next seven columns give the amount of changes in the medium-sized arteries. For convenience the sections are numbered from 1 to 7. The next seven columns of the table list the microscopic changes in the smallest arteries and the arterioles, which are listed together because they are considered a functional unit. In grading all the arteries and arterioles, the degree of sclerosis was considered from the point of view of the amount of narrowing of the lumen of the vessel. Four plus sclerosis was taken to indicate complete occlusion, and three plus almost complete occlusion.

In the next seven columns the amount of interstitial fibrosis is given for all the sections. It will be noticed that in very few sections was there a complete lack of fibrosis. None of the hearts showed a total lack of fibrosis. Examples of fibrosis are shown in figures 1 and 2.

## SUMMARY OF OBSERVATIONS

The observations which attract most attention are the amount and distribution of the myocardial fibrosis and the relation of the latter to the arterial changes. Clawson<sup>13</sup> and Hunter<sup>14</sup> recently claimed that the fibrosis is proportional to the amount of sclerosis in the coronary arteries. Examination of the table will quickly show that this is not the case here. Only 6 hearts had marked sclerosis of the large coronary arteries; 4 had moderate, and 17 slight, sclerosis. Only 1 heart had marked sclerosis of the medium-sized arteries (those too small to be opened with a pair of scissors and less than the diameter of a low power field of 100 magnifications), and 8 had slight changes in these vessels. In the remaining 18 hearts no changes could be discerned in the medium-sized arteries. On examining the smallest arteries and arterioles, it was observed that 6 hearts had marked arteriolosclerosis in one or more sections (fig. 1). 13 had a slight and insignificant degree of arteriosclerosis and 8 had no changes in these vessels.

Only those cases with marked sclerosis in the arterial tree were considered to show enough anatomic evidence of an interference with

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14. Hunter, O. B., quoted by Brown.<sup>9</sup>

TABLE 1.—Main Clinical Data and Gross Cardiac

Case	Race	Sex	Age	Blood Pressure	Weight of Heart, Gm.	Thickness of Left Ventricle, Mm.	Thickness of Right Ventricle, Mm.	Related Myocardial or Vascular Changes	Cause of Death	Degree of Sclerosis of Large Coronary Arteries
1	W	M	73	220/160	650	20	4	Left ventricle thinned to 3 mm. at apex, mural thrombi in left ventricle, myomalacia in region of left papillary muscle	Cardiac failure	Marked
2	C	M	33	180/130	650	18	6	Left ventricle thinned to 3 mm. at apex, mural thrombi in left ventricle	Cardiac failure	Slight
3	C	M	64	265/158	580	20	3	Left ventricle thinned to 3 mm. at apex, bundle branch block clinically (verified microscopically)	Cardiac failure	Slight
4	C	M	67	184/104	550	20	5	Calcification of aortic ring, dissecting aneurysm of aorta	Spontaneous rupture of aorta	Marked
5	C	M	46	160/125	705	19	7	Moderate atheroma of the aorta, syphilitic aortitis	Cardiac failure	Marked
6	W	F	72	.....	570	25	5	Marked atherosclerosis of aorta, syphilitic aortitis with aneurysm of arch	Cardiac failure	Slight
7	C	F	29	274/168	530	25	5	Chronic glomerulonephritis .....	Renal insufficiency (uremia)	Slight
8	W	M	53	190/140	630	15	3	Mural thrombi in left ventricle....	Cardiac failure	Slight
9	W	M	88	170/110	475	17	6	Marked sclerosis of mitral ring, sclerosis at aortic ring, aneurysm of left ventricle, thrombi in left iliac vein and right pulmonary artery	Cardiac failure	Slight
10	W	M	54	260/152	720	30	5	Ostium of left coronary artery narrowed	Cardiac failure	Marked
11	W	M	46	260/154	490	20	3	Anatomically patent foramen ovale	Cerebral hemorrhage	Moderate
12	C	M	66	250/160	775	25	7	Syphilitic aortitis .....	Cardiac failure, pernicious anemia	Slight
13	W	M	46	180/136	550	20	4	Fibrinous pericarditis .....	Cardiac failure	Slight
14	C	M	81	.....	590	20	5	.....	Cardiac failure	Slight
15	C	M	45	170/110	480	14	5	Very severe coronary sclerosis with complete occlusion of both arteries, thrombotic occlusion of left posterior cerebral artery	Severe coronary sclerosis, encephalomalacia from occlusion of left posterior cerebral artery	Marked
16	W	M	60	220/126	940	26	11	Subendothelial hemorrhages .....	Cardiac failure	Moderate
17	W	F	58	.....	645	21	5	Slight fibroplastic deformity of mitral and aortic valves .....	Cardiac failure	Slight
18	W	M	50	240/170	565	23	3	.....	Generalized peritonitis following resection of bowel	Slight
19	W	F	42	262/160	605	19	9	Slight fibroplastic deformity of mitral valve .....	Cardiac failure	Slight
20	C	F	42	242/138	735	20	6	.....	Cerebral hemorrhage	Marked
21	W	M	63	190/120	500	19	4	Hemorrhage in cerebellum.....	Renal insufficiency (uremia)	Slight
22	C	F	45	270/180	540	20	4	Malignant nephrosclerosis .....	Cardiac failure	Moderate
23	C	M	65	115/ 85	590	16	6	Multiple anthracotic—silicotic nodules in lungs	Renal insufficiency (uremia)	Slight
24	C	F	40	244/170	515	28	5	Malignant nephrosclerosis .....	Cardiac failure	Moderate
25	W	M	54	165/ 60	740	22	8	Recent cortical encephalomalacia..	Cardiac failure	Slight
26	W	F	63	140/ 90	700	18	4	Mural thrombus in left ventricle, fibrinopurulent pericarditis	Cardiac failure, small cortical fibrosarcoma of left kidney	Slight
27	C	M	48	184/112	615	20	4	Mural thrombi in left ventricle and right auricle, thrombi in pulmonary artery, prostatic abscesses with emboli	Cardiac failure, prostatic abscess with emboli	Slight

### *Findings in Twenty-Seven Hypertensive Hearts*

the blood supply to have caused the myocardial fibrosis. The 13 cases in the category just cited overlap somewhat because the heart with marked sclerosis of the medium-sized arteries and 1 of the 6 with marked sclerosis of the arterioles also had marked sclerosis of the large coronary arteries. This leaves 11 hearts with marked anatomic changes in the arterial tree. In the remaining 16 cases the moderate

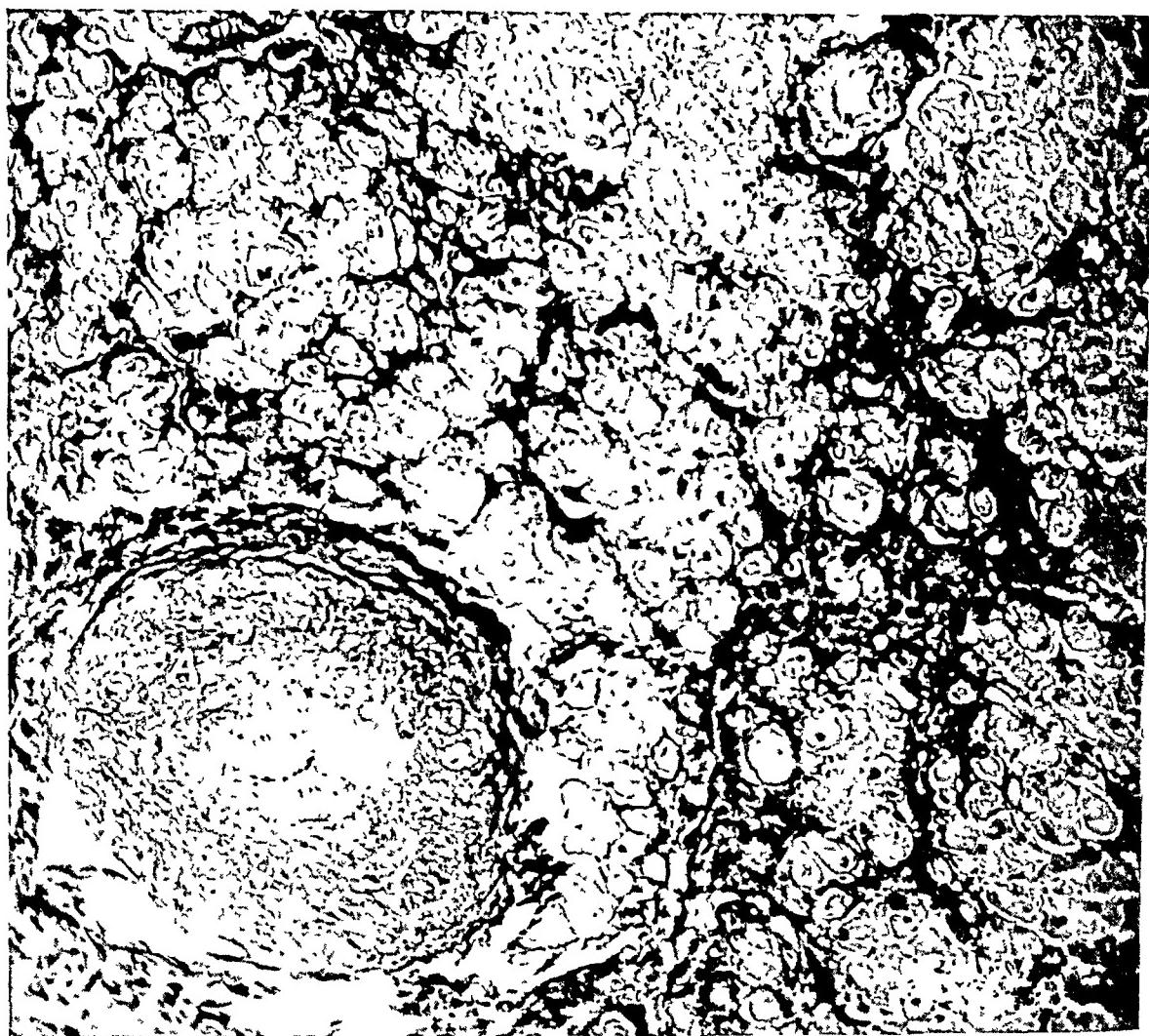


Fig. 1 (case 23).—Section showing diffuse scattered scarring in an area with marked arteriosclerosis (moderate sclerosis of the large coronary arteries). *A* indicates a much thickened arteriole; *B*, slightly thickened arteriole, and *S*, the scar. van Gieson's stain;  $\times 150$ .

or slight changes in the large vessels and the amount of sclerosis in the smaller vessels (classed as one or two plus, at the most) were considered insufficient to have caused the microscopic fibrosis of the myocardium. The fibrosis in this second group must, then, be due to functional interference with the blood supply or to some other factor.

Other factors considered have been the weight of the heart, race, sex, age, blood pressure, the cause of death and the relation of the condition to syphilis. The hearts varied in weight from 475 to 940 Gm.; 3 weighed less than 500 Gm., 11 weighed between 500 and 600, 6 weighed from 600 to 700, 6 from 700 to 800 and 1 more than 900 Gm. The grouping of race and sex gives 10 white males, 9 colored males, 4

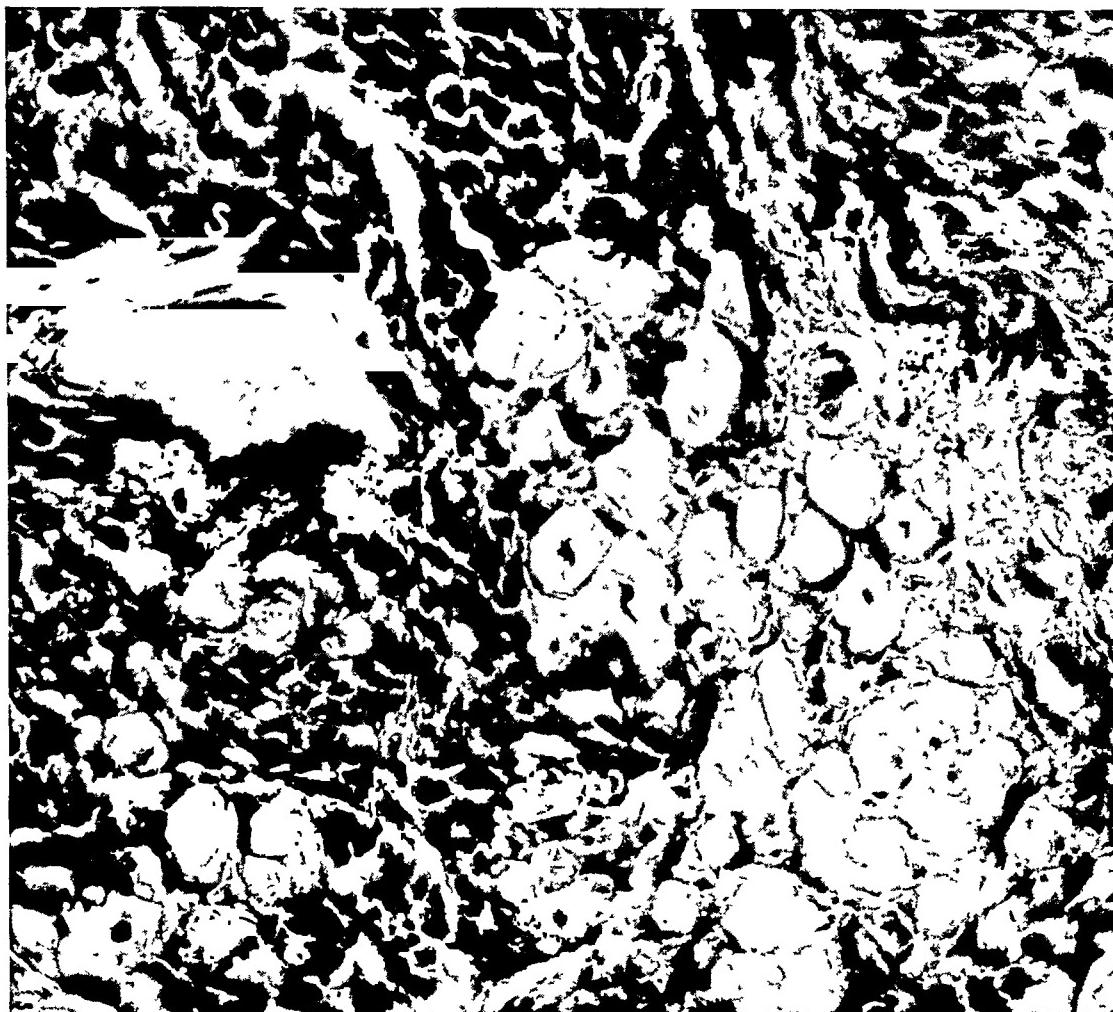


Fig. 2 (case 16).—Section showing a large interstitial scar. There was no arteriosclerosis or arteriolosclerosis in the heart. The scar tissue has surrounded and separated the muscle fibers and caused atrophy. *S* indicates the scar; *N*, normal-sized muscle fibers, and *A*, atrophic muscle fibers van Gieson's stain;  $\times 300$ .

white females and 4 colored females. The ages varied from 29 to 81, 1 patient being under 30, 9 between 30 and 40, 6 between 40 and 50, 7 between 50 and 60, 1 between 70 and 80, and 2 over 80. No correlation could be established between these factors and the myocardial fibrosis.

Dogliotti<sup>15</sup> and Miller and Perkins<sup>16</sup> recently showed that myocardial fibrosis does not develop as a result of advancing age. Thus the age of the patients could have no bearing on the amount of myocardial fibrosis present.

As to the blood pressure, in all but 2 of the cases (23 and 26) in which the blood pressure is given there was a definite hypertension. These 2 patients undoubtedly had a previous hypertension, since, as Willius and Smith<sup>17</sup> pointed out, hypertension may vanish, leaving only an enlarged heart.

With respect to the cause of death the cases can be divided into two main groups—19 cases in which death was due to cardiac failure and 7 in which it was due to other causes. The second group consists of 3 cases (7, 22 and 24) in which death was caused by renal insufficiency, 2 (11 and 21) in which it was due to accidents to the cerebral vessels, 1 (4) in which it resulted from ruptured dissecting aortic aneurysm and 1 (15) in which it was caused by occlusions of the cerebral and coronary arteries. In the remaining case (18) the patient died of peritonitis.

The factors of blood pressure and cause of death bore no noticeable relationship to the gross and microscopic observations. It is especially interesting, but not surprising, that no definite histologic change can be demonstrated after myocardial failure, since the shift from a state of compensation to one of decompensation is mainly an alteration of functional efficiency.

Syphilitic aortitis was present in only 3 cases (5, 6 and 12). No correlation can be demonstrated between the presence of syphilis and the amount of myocardial scarring present. Since the percentage of syphilis in this small series is approximately equal to the 10.3 per cent of syphilitic aortitis which Jaffé<sup>18</sup> found in 3,300 autopsies at the Cook County Hospital, it is reasonable to suppose that none of the other patients in my group had syphilis. For this reason the myocardial scars are not believed to have any relation to syphilis, and they are not similar to the syphilitic scars which Warthin<sup>19</sup> reported, because of the absence of any infiltrative phenomena in the region of the scars.

At this point it may be mentioned that stretching of the myocardium could not have caused the myocardial fibrosis, because the fibrosis was also observed in patients who had not died of decompensation of the heart, and because it was not the uniform, diffuse, perimuscular fibrosis that occurs with experimental dilatation of the ventricles.

15. Dogliotti, G. C.: Ztschr. f. d. ges. Anat. (Abt. 1) **96**:680, 1931; Verhandl. d. anat. Gesellsch. **71**:14, 1931.

16. Miller, A. M., and Perkins, O. C.: Am. J. Anat. **39**:205, 1927.

17. Willius, F. A., and Smith, H. L.: Am. Heart J. **8**:170, 1932.

18. Jaffé, R. H.: Klin. Wchnschr. **10**:2081, 1931.

19. Warthin, A. S.: Proc. Inst. Med., Chicago **8**:173, 1931.

Endocardial sclerosis was also present in every heart. It is interesting that this sclerosis was, as a rule, most marked in the left auricle, the chamber which had the least amount of myocardial fibrosis. The most marked myocardial scarring was observed, on the other hand, in the left ventricle, especially in the posterior wall and the large anterior papillary muscle. The elastic tissue stain and van Gieson's stain revealed

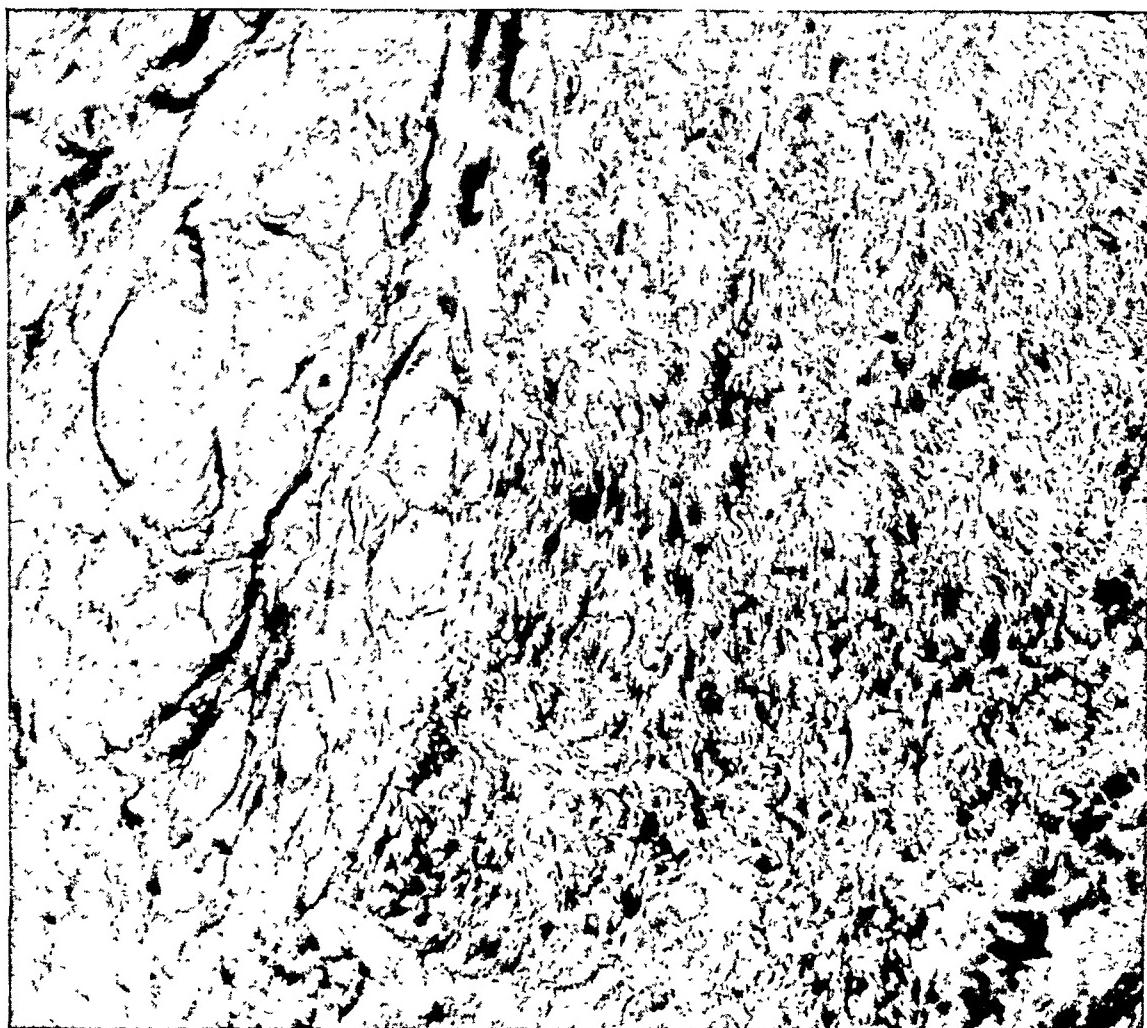


Fig. 3 (case 16).—Same scar as in figure 2, showing the large amount of elastic fibrils. *M* indicates the myocardium, and *S*, the scar. Weigert's elastic tissue stain;  $\times 300$ .

that the endocardial thickening was due to an increase in the number of smooth muscle cells and elastic fibers normally present in the sub-endocardial layers. The elastic tissue stain also showed that the myocardial scars were composed of elastic fibrils varying from 30 to 100 per cent in various scars (fig. 3). As Perkins and Miller<sup>20</sup> pointed

20. Perkins, O. C., and Miller, A. M.: Arch. Path. 3:785, 1927.

out, collagenic connective tissue can be converted into elastic connective tissue under the influence of stress and strain, contrary to the general notion that newly formed elastic tissue must be derived from preexisting elastic fibers.

#### COMMENT

Many recent studies on the hypertensive heart and fibrosis of the myocardium (Brown,<sup>9</sup> Cowan,<sup>21</sup> Barnes and Ball,<sup>22</sup> Burton and his collaborators,<sup>23</sup> and Gilchrist and Ritchie<sup>24</sup>) have failed to describe adequately the microscopic changes. Two recent articles, however, give microscopic observations in some detail. Clawson<sup>13</sup> studied a series of 139 hypertensive hearts, making microscopic examinations at five different places in each heart. These hearts were divided into four groups, in which death was due to severe coronary sclerosis, cardiac failure, cerebral hemorrhage or renal insufficiency. Clawson stated that the amount of myocardial fibrosis was proportional to the degree of coronary sclerosis. His figures showed that the hearts in the group in which death was due to coronary sclerosis had the most marked amount of myocardial fibrosis, but in the other groups the relationship was not proportional. Similarly, in my series, the most marked degree of myocardial fibrosis was present in the group with marked coronary sclerosis, but many hearts with only slight changes in the arterial tree had an amount of myocardial scarring equal to or greater than that in some of the hearts with marked coronary sclerosis.

Fitz Hugh<sup>6</sup> examined grossly and microscopically 228 hearts chosen on the basis of hypertrophy or fibrosis. Fourteen of these hearts he studied in great detail, making sections from six different locations. He concluded that while thick coronary arteries and marked myocardial fibrosis often occurred together, it was not unusual to find them present independently; the same was true as regards the relation of arteriosclerosis and myocardial fibrosis. These views are thus contrary to those of Clawson. Mönckeberg,<sup>25</sup> too, in discussing fibrosis of the myocardium, stated that a definite parallelism between the grade of fibrosis and the severity of coronary sclerosis cannot be found.

Perkins and Miller,<sup>20</sup> studying a group of hypertensive hearts, observed changes similar to those in the group of 27 just described. They observed a much thickened endocardium, and many newly formed

21. Cowan, J.: *Lancet* **2**:1, 1930.

22. Barnes, A. R., and Ball, R. G.: *Am. J. M. Sc.* **183**:215, 1932.

23. Burton, J. A. G.; Cowan, J.; Kay, J. H.; Marshall, A. J.; Rennie, J. K.; Ramage, J. H., and Teacher, J. H.: *Quart. J. Med.* **23**:293, 1930.

24. Gilchrist, A. R., and Ritchie, W. T.: *Quart. J. Med.* **23**:273, 1930.

25. Mönckeberg, J. G.: *Die Erkrankungen des Myokards und des spezifischen Muskelsystems*, in Henke, F., and Lubarsch, O.: *Handbuch der speziellen pathologischen Anatomie und Histologie*, Berlin, Julius Springer, 1924, vol. 2.

elastic fibers and numerous scars throughout the myocardium. Their description of the scars as islands of connective tissue which enclosed fragments of degenerating muscle fibers exactly fits the picture seen in most of my sections. One obtains the impression that there has been a gradual shrinking of the muscle fibers, with replacement by fibrous tissue. Perkins and Miller believed that these scars were caused by a decreased nutritive supply. Decrease of the blood supply was also given by Cohn<sup>25</sup> as the cause of the scars which he described in the myocardium.

Other authors, too, have believed that an adequate or inadequate blood supply is the factor determining the myocardial changes. Miller and Weiss<sup>26</sup> studied diseases of the coronary arteries and concluded that adequate collateral circulation explained the lack of hypertrophy of the heart in the presence of coronary sclerosis. Jaffé and Bross<sup>27</sup> stated that the number of anastomoses between the coronary arteries and not the degree of occlusion determined the amount of myocardial damage. Christian<sup>28</sup> believed that hypertrophy of the heart often develops to such an extent that the capillaries cannot supply the muscle with sufficient blood, thus establishing a vicious cycle leading to decompensation and death.

Even in large macroscopic infarcts or areas of fibrosis it is not always possible to find definite occlusion of the large coronary arteries. Brown<sup>29</sup> found 25 hearts with normal coronary arteries among 110 hearts with gross fibrosis. In a series of 49 hearts with old infarcts, reported by Barnes and Ball,<sup>22</sup> many did not have complete coronary occlusion. Levine and Brown<sup>29</sup> reported 46 cases in which the heart showed large infarcts or infarct scars, and failed to find definite coronary occlusion in 11. Lisa and Ring<sup>30</sup> found normal coronary arteries in 8 hearts of a series of 100 with well circumscribed gross infarction or fibrosis. They could locate definite occlusion in only 57. Krumbhaar and Crowell<sup>31</sup> made an extensive report of spontaneous rupture of the heart and concluded that changes in the coronary arteries could not explain all cases of spontaneous rupture. Davenport<sup>32</sup> reported 52 cases of rupture of the left ventricle; he found changes in the coronary arteries in only 30. He concluded that more detailed

26. Miller, H. R., and Weiss, W. M.: Arch. Int. Med. **42**:74, 1928.

27. Jaffé, Rudolf, and Bross, K.: Centralbl. f. allg. Path. u. path. Anat. **56**: 246, 1933; Ztschr. f. klin. Med. **123**:63, 1933.

28. Christian, H. A.: South. M. J. **20**:28, 1927.

29. Levine, S. A., and Brown, C. L.: Medicine **8**:245, 1929.

30. Lisa, J. R., and Ring, A.: Arch. Int. Med. **50**:131, 1932.

31. Krumbhaar, E. B., and Crowell, C.: Am. J. M. Sc. **170**:828, 1925.

32. Davenport, A. B.: Am. J. M. Sc. **176**:62, 1928.

microscopic studies were necessary to arrive at any definite solution of this problem.

Such a careful study has recently been made by Jaffé and Bross.<sup>27</sup> These authors reported 9 cases in all, in 7 of which death was due to spontaneous rupture of the heart. In the first 2 cases the rupture occurred suddenly, the patients being in good health previously. The coronary vessels were smooth and unobstructed. In both these cases careful microscopic examination showed widespread changes both at, and some distance from, the rupture. These changes consisted of multiple focal areas of necrosis interspersed with areas containing marked engorgement of the capillaries. Jaffé and Bross concluded that functional disturbances in the terminal arterial tree led to the multiple areas of necrosis and the cardiac rupture.

In their next 3 cases the gross and microscopic observations were identical, but in addition in each instance there was an occluding thrombus in a branch of the coronary artery leading to the area of rupture. In the next 2 cases there was marked coronary sclerosis without definite occlusion. Yet all 5 of these hearts had the same microscopic areas of necrosis, leukodiapedesis and leukostasis present in their first 2 cases. These changes were much too diffuse and severe to have been accounted for by the condition observed in the coronary arteries, especially in the 3 hearts in which coronary occlusion was the only finding. In some areas older changes, such as fibrosis, were visible grossly and microscopically.

The last 2 hearts which Jaffé and Bross included in their series did not have a rupture. The first had an incomplete tear and marked coronary sclerosis. The second (their case 9) had widespread fibrosis, which was visible grossly, but no coronary changes. In both these hearts careful histologic study revealed numerous recent changes (necrosis, extravasation of leukocytes and stasis) similar to those in their first 7 cases, even in the areas of marked scarring.

In addition, Jaffé and Bross mentioned similar findings and observations of other investigators. Büchner<sup>33</sup> observed identical areas of myocardial necrosis in 5 grossly normal hearts from patients with angina pectoris. Oberndorfer<sup>34</sup> observed changes in the cardiac muscle in cases of angina pectoris similar to those in cases of sclerosis of the coronary arteries and believed their cause to be vascular spasm. Wollheim<sup>35</sup> reported a case in which the diagnosis was cardiac infarct and in which the only finding was subepicardial hemorrhage, due, he thought, to vascular spasm. Gruber and Lanz<sup>36</sup> found areas of "ischemic necrosis"

33. Büchner, F.: Beitr. z. path. Anat. u. z. allg. Path. **89**:644, 1932.

34. Oberndorfer: München. med. Wchnschr. **72**:1495, 1925.

35. Wollheim, E.: Deutsche med. Wchnschr. **57**:617, 1931.

36. Gruber, G. P., and Lanz, H. F.: Arch. f. Psychiat. **61**:98, 1920.

in a patient with epilepsy, and Neubürger<sup>37</sup> reported several cases of epilepsy in which the hearts had smooth coronary arteries and areas of "ischemic necrosis" of the myocardium, intermingled with scars. He attributed the changes to spasm of the vessels.

Other authors (Mönckeberg,<sup>25</sup> Levine and Brown,<sup>29</sup> Smith and Bartels<sup>38</sup> and Buckley<sup>39</sup>) noted similar myocardial changes, but failed to give them the same significance, usually considering them inflammatory. Jaffé and Bross<sup>27</sup> and Büchner<sup>33</sup> carefully examined large infarcts similar to those reported by Levine and Brown. Despite an obvious adjacent coronary occlusion they found that these infarcts were not uniform, as was suggested by their gross appearance, but were multiple, confluent small areas of necrosis.

In order to understand the underlying cause of the changes which Jaffé and Bross reported, it is necessary to refer to the studies of Ricker<sup>40</sup> and his school. Briefly stated, Ricker found that the blood vessels react in segments to stimuli brought to them, each segment being a functional unit. The large vessels react differently from the medium-sized ones and from the smallest vessels (small arteries, arterioles and capillaries, which form a functional unit). The terminal vascular segments (which Ricker believed to receive a great number of nervous impulses) react to weak irritation by vasodilatation, to medium irritation by vasoconstriction and to strong irritation by marked vasodilatation and slowing of the blood stream, while the proximal arterial segments remain constricted. This is the state which Ricker calls "peristasis" (inflammatory hyperemia), which leads without sharp demarcation to "prestasis" (vasoparesis) and, in the last degree, to stagnation of the blood stream, or "stasis" (vasoparalysis). Peristasis causes exudation of serum and leukocytes; prestasis causes irregularity in the blood contents and diapedesis of erythrocytes, and stasis causes stoppage of blood flow and conglutination of red blood cells, which are no longer individually recognizable.

According to Ricker, in the earlier stages the terminal arterioles may be constricted, while the capillaries become dilated. Thus the blood stream has a lessened *vis a tergo* and becomes slowed in the capillaries. Later the arterioles dilate, while the more proximal arterial segments remain constricted, resulting in a similar slowing of the blood stream in the arterioles. This leads to the changes mentioned and finally ends in stoppage of the blood stream in stasis. Even when stasis is reached

37. Neubürger, K.: Verhandl. d. deutsch. path. Gesellsch. **23**:487, 1928; Klin. Wehnschr. **12**:1355, 1933.

38. Smith, H. L., and Bartels, E. C.: M. Clin. North America **15**:1585, 1932.

39. Buckley, R. C.: Am. J. Path. **4**:249, 1928.

40. Ricker, G.: Pathologie als Naturwissenschaft- Relationspathologie, Berlin, Julius Springer, 1924.

the process is not irreversible, and a return to normal is possible if the exciting factor, i. e., the nervous stimulus, is removed. Also, this sequence of events is not always carried through gradually in all its stages, but its speed may vary considerably at any one stage. Necrosis may and frequently does follow stasis, when the stasis persists and becomes permanent. Ricker stated that necrosis may follow either organic blocking of a vessel or stasis, each of which severs the connection between part of an organ and the blood stream.

According to Jaffé and Bross, Ricker's views, which are supported by his experimental work and partly by that of Tannenberg,<sup>41</sup> explain the microscopic observations of necrosis, leukodiapedesis and stasis in their series of hearts and in the cases which they cited. They advanced the suggestion that thinning over an occluded point or dilatation of the coronary artery ahead of an occlusion may send the stimuli to the terminal vessels and lead to the changes described. When there is no occlusion they believe that spasm of a large artery may supply the needed stimulus. Thus, Jaffé and Bross believe that the widespread small necrotic changes are not "ischemic" but "static."

It must be mentioned, however, that not all recent investigators agree entirely with Ricker. Fischer-Wasels,<sup>42</sup> in a recent review of the subject, came to the conclusion that a toxic factor must also be present to cause necrosis. Peissachowitsch<sup>43</sup> recently disproved some of Ricker's ideas but not his underlying thesis.

Pathologic alterations of other organs have also been explained on the basis of Ricker's theory. Shapiro<sup>44</sup> applied the theory to the changes in the kidneys in malignant nephrosclerosis. Scrivener and Oertel<sup>45</sup> and Wolfson<sup>46</sup> used it to explain bilateral cortical necrosis of the kidneys in pregnancy, and Ricker<sup>40</sup> and his associates explained various pathologic conditions on the basis of functional vascular disturbances.

As Neubürger<sup>47</sup> and Jaffé and Bross<sup>27</sup> (especially in their case 9) pointed out, the late severe necrotic changes may be intermingled with older interstitial areas of fibrosis. It is easily conceivable that in some hearts only the fibrosis may be present, owing to the slower development or minor severity of the circulatory disturbances.

This seems to hold true for the 16 hearts without organic vascular changes in my series of 27 hypertrophic hearts. All 16 had scattered microscopic areas of fibrosis. The microscopic picture of the areas gives

41. Tannenberg, J.: Frankfurt. Ztschr. f. Path. **31**:173, 1925.

42. Fischer-Wasels, B.: Frankfurt. Ztschr. f. Path. **45**:1, 1933.

43. Peissachowitsch, K.: Frankfurt. Ztschr. f. Path. **45**:246, 1933.

44. Shapiro, P. F.: Arch. Int. Med. **48**:199, 1931.

45. Scrivener, W. de M., and Oertel, H.: J. Path. & Bact. **33**:1071, 1930.

46. Wolfson, A.: Tr. Chicago Path. Soc. **14**:33, 1932; personal communication to the author.

one the impression that there has been a gradual melting away of the muscle fibers without acute necrosis (which would be the change in the analogous rapid process) and a gradual replacement of this disappearing muscle tissue by scar tissue.

The observations for these hearts are comparable to those in benign nephrosclerosis, and the observations in case 9 of Jaffé and Bross resemble those in severe rapidly developing necrotic lesions of malignant nephrosclerosis engrafted on the changes of benign arteriolosclerosis of the kidneys. The changes in the first 2 cases which Jaffé and Bross reported are analogous to those in malignant nephrosclerosis without any preexisting benign sclerotic vascular changes. In my series 11 hearts had organic vascular changes which, by interfering with the blood supply, might account for the scarring, but which, in accordance with the statement of Jaffé and Bross, may also have been due, at least in part, to spasm and later paralysis of the terminal arterial segments.

#### SUMMARY

Twenty-seven hypertensive hearts were examined grossly and microscopically. For the microscopic examination blocks of tissue were taken from seven different locations in each heart.

Six hearts were found to have marked sclerosis of the large coronary arteries. Six hearts had a marked sclerosis of the arterioles in one or more sections. Since 1 of the 6 with marked arteriolosclerosis also had marked sclerosis of the large coronary arteries, there were 11 hearts which had marked organic changes in the arterial tree. The remaining 16 hearts had changes in the arterial tree which were considered too slight to have seriously interfered with the blood supply.

In all the hearts minute myocardial scars were observed in many areas. As a rule, this fibrosis was most marked in the left ventricle. The most marked scarring was present in the hearts with marked alteration of the wall of the arteries and arterioles, but many hearts in the group without organic arterial changes also had marked interstitial fibrosis.

No correlation could be found between age, sex, race, weight of the heart, blood pressure, the cause of death or the presence of syphilis and the amount of myocardial fibrosis. Many of the scars in the myocardium were observed to contain varying amounts of elastic tissue. This bears out the contention of Perkins and Miller that stress and strain can convert collagenic connective tissue into elastic tissue.

Endocardial sclerosis was present in all the hearts, being as a rule, most marked in the left auricle. It was usually due to an increase in smooth muscle cells and elastic fibers.

Failing to find an anatomic basis for the myocardial fibrosis present in the 16 hearts without organic changes in the arterial tree, one is forced to rely on an explanation of functional disturbances. The views of Ricker on functional spasm and later vasoparalysis of the terminal arterioles, interpreted in the light of the work of Jaffé and Bross, appear to give a logical solution of the question. In these 16 hearts the functional changes undoubtedly acted over a long period and in a mild degree, leading only to myocardial fibrosis.

# BILATERAL GLIOBLASTOMA MULTIFORME

REPORT OF TWO CASES

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A glioblastoma multiforme which extends across the midline to involve adjacent brain tissue of the opposite hemisphere is not particularly rare.<sup>1</sup> However, glioblastoma multiforme appearing in widely separated centers in the two hemispheres is quite unusual.<sup>2</sup> Because of this fact the following report of two cases presenting that condition seems warranted.

The cases studied appeared in a series of forty autopsies on patients with tumors of the brain. They represent the only bilateral growths in this series with the exception of one case of bilateral perineural fibroblastoma of the auditory nerves.

When first studied both were thought to be cases of bilateral glioblastoma multiforme of multicentric origin. A careful study of sections proved that in case 1 the growth was an extension from the left temporal to the right parietal cortex. It was represented only by a fine filamentous tumor tissue near the midline. In case 2 the tumor is considered a bilateral tumor of multicentric origin. No communication whatever could be demonstrated between the two growths, and each appeared as a separate, well defined tumor.

The possibility of one of these growths (case 2) being a metastasis from the other cannot be entirely ruled out since each growth came into contact with a small area of the arachnoid. Also, the right tumor formed a portion of the wall of the right ventricle, thus predisposing to metastasis.<sup>3</sup> While this possibility is fully admitted, it is not considered the most likely explanation because of the character of these two lesions. Each tumor appeared as a separate entity with no metastases or extensions into the subarachnoid space.

An interesting feature is the fact that in each case one tumor conformed quite closely to the "typical" glioblastoma multiforme.<sup>4</sup> while

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From the Department of Bacteriology and Pathology, Indiana University School of Medicine, aided by a grant from the Research Division.

1. Bailey, Percival: *Intracranial Tumors*, Springfield, Ill., Charles C. Thomas, Publisher, 1933.

2. Hosoi, K.: *Arch. Neurol. & Psychiat.* **24**:311, 1930. Penfield, Wilder: Personal communication to the author.

3. Cairns, H., and Russel, D. S.: *Brain* **54**:377, 1931.

4. Deery, E. M.: *Bull. Neurol. Inst. New York* **2**:157, 1932.

that of the opposite hemisphere was distinctly of the "astrocyte type" of glioblastoma multiforme.

No attempt has been made to correlate these cases with multiple gliomas previously reported. Such a summary may be left until more complete records are available and more uniform methods and terminology come into general use.

#### REPORT OF CASES

CASE 1.—A white man, aged 48, entered the Robert W. Long Hospital on Sept. 26, 1932, complaining of frontal and occipital headaches of four months' duration. The headaches had become quite severe during the latter months of his illness and were most intense in the right occipital region, radiating to the vertex. Occasional vomiting became a feature of the illness two weeks before death. Pertinent physical findings included impairment of memory, bilateral optic edema and a spinal fluid pressure of 20 mm. of mercury with a normal Queckenstedt sign. The left biceps, triceps and ankle jerks were absent. On the right the deep reflexes and the left knee jerk were normal. There was no alteration in muscular strength or cutaneous sensation. A diagnosis of right parietal tumor of the brain was made. A right parietal decompression was done, and an inoperable tumor was found. The patient died thirteen days after the operation.

*Gross and Microscopic Examination of the Brain.*—The meningeal vessels were moderately injected and the convolutions flattened. The ventricles were dilated, and a few small cysts (1 or 2 mm.) were found in the choroid plexus of the right ventricle. There was no evidence of metastasis other than that to be described.

Left Hemisphere (fig. 1): Over the left posterior part of the temporal lobe, just below the superior temporal sulcus, was a large area of purplish-brown cortex. This consisted in the outer portion of a soft, degenerated, brownish tumor, measuring 3 by 4 by 7 cm. The cortex was involved to a lesser extent than the subcortex. The tumor contained numerous small hemorrhages, and its margins were indefinite with no attempt at encapsulation. The growth extended to the posterior horn of the lateral ventricle where it made up a sizable portion of the wall of that structure. From the left lateral ventricle the tumor appeared to extend medially by a thin pedicle growth. This continued behind the splenium of the corpus callosum across the midline and into the right hemisphere. There it formed the lesion on the right side, to be described.

*Microscopic Examination:* The tumor consisted of a vascular and cellular tissue. The vessels presented an endothelial and adventitial overgrowth, of which the latter was more prominent. Neuro-ectodermal cells were numerous and were found in clumps and also diffusely distributed throughout the section. They joined with the spongioblasts to form a pseudopalisade about many degenerating and necrotic areas. The spongioblasts were mostly of the apolar type and were well demonstrated with phosphotungstic acid and hematoxylin. A few multinucleated giant cells were noted, and mitotic figures were not infrequent. Many astrocytes fringed the tumor tissue. When stained with gold chloride these showed varying degrees of degeneration. Only comparatively few astrocytes were found in the tumor itself. This was made up principally of spongioblasts and neuro-ectodermal cells as stated.

The diagnosis was glioblastoma multiforme of the left temporal lobe.

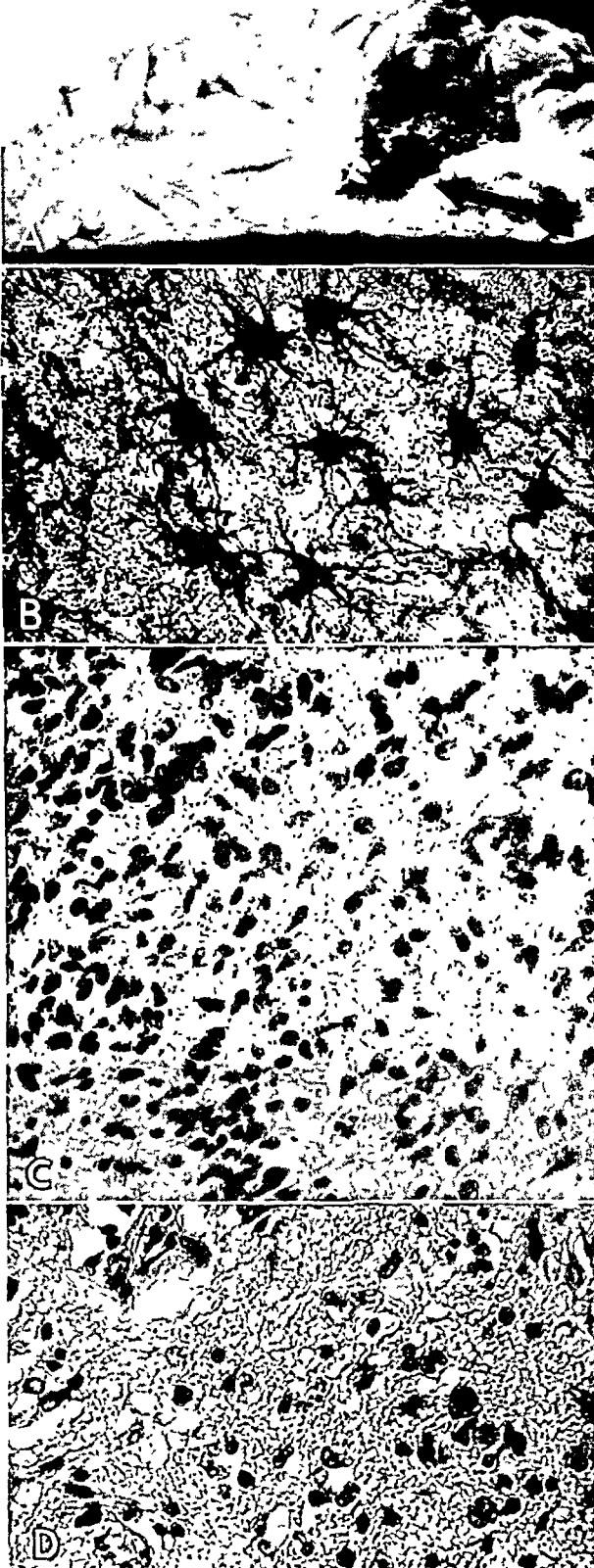


Fig. 1 (case 1).—*A*, tumor of the left temporal lobe forming part of the left lateral ventricle indicated by the arrow; *B*, varying degrees of astrocyte degeneration fringing the neoplasm (gold chloride); *C*, general structure, slight palisading, occasional mitotic figures and adventitial proliferation (hematoxylin and eosin); *D*, area of tumor containing many astrocytes (phosphotungstic acid and hematoxylin). Reduced from  $\times 450$  and  $\times 300$ .

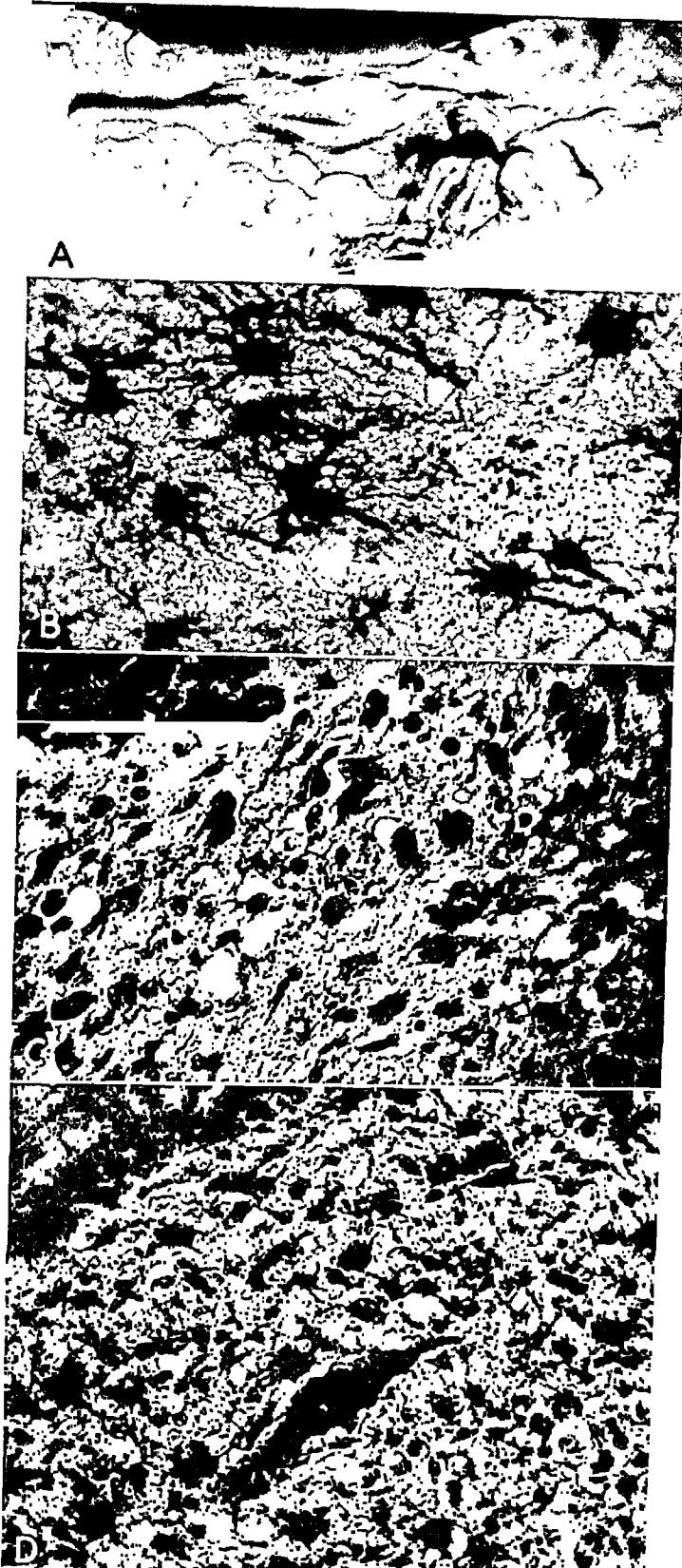


Fig. 2 (case 1).—*A*, right parietotemporal tumor; *B*, degenerating astrocytes in the marginal zone (gold chloride); *C*, representative area made up largely of astrocytes (hematoxylin and eosin); *D*, astrocytes, spongioblasts and neuro-ectodermal cells in the subarachnoid space. Sucker feet attached to the thickened vessel wall are poorly shown (phosphotungstic acid and hematoxylin). Reduced from  $\times 450$  and  $\times 300$ .

Right Hemisphere (fig. 2) : In the parietotemporal region just posterior to the postcentral gyrus was a cortical area of degeneration and hemorrhage. This measured 3 cm. in diameter and was roughly pyramidal in shape (fig. 2). The apex extended medially around the ventricle to join the previously noted small pedicle of tumor tissue from the opposite hemisphere. Grossly and microscopically the tumor did not involve the right ventricle, although at one point the wall of the ventricle was partially necrotic. The bulk of the tumor was deep in the white matter and peripherally involved the sulci, leaving several of the gyri relatively intact (fig. 2). The tumor tissue was much more firm and limited than that of the opposite side. Degeneration was less on the right, though there were numerous hemorrhages.

Microscopic Examination: Sections of the tumor tissue showed it to be invading the subarachnoid space. In contradistinction to the opposite side, the astrocytes were numerous and prominent while there were fewer spongioblasts. Occasional multinuclear giant cells were found, and there were a few mitotic figures. There were many neuro-ectodermal cells scattered throughout the section, and clumps of them were found around degenerating areas. Portions of the tumor had a loose structure with the predominance of astrocytes suggesting an astrocytoma. The most striking feature of the tissue was the extreme thickening of the walls of the numerous blood vessels. This consisted for the most part of an endothelial proliferation, but hyperplasia of the adventitia was also found. Some of the vessels were completely occluded by the process. Section through the right ventricle showed the wall of that structure to be involved by the tumor.

The diagnosis was glioblastoma multiforme of the right parietal cortex by direct extension from the glioblastoma multiforme of the left temporal lobe.

*Comment.*—This case was considered one of multiple glioma in which the tumor of the left temporal region was primary. This neoplasm, glioblastoma multiforme, extended by a thin filamentous growth across the midline into the right parietal cortex where a second similar neoplasm was produced.

CASE 2.—An American housewife, aged 41, entered the Robert W. Long Hospital on June 21, 1933, complaining of drowsiness of three weeks' duration. Her lethargy had been slowly but steadily increasing, and on admission she could be aroused only with difficulty. Since the onset the patient had eaten but little, and constipation had been quite severe. A routine physical examination revealed only slightly hyperactive ankle and biceps reflexes. These were equal for the right and left sides. The pupils were sluggish but reacted equally to the light stimulus. The laboratory examinations showed a white cell count of 12,000 with 90 per cent polymorphonuclears. The blood sugar, total nonprotein nitrogen and red cell count were within normal limits. The urine was loaded with pus cells, and for this reason immediate attention was focused on clearing up a rather severe pyelitis. Occasional coarse tremors of the arms and legs were noted after admission. The patient died suddenly on the first day after entering the hospital before a neurologic study could be made.

*Gross and Microscopic Examination of the Brain* (fig. 3).—The vessels over the vertex were engorged and prominent. There was no dilatation of the ventricles. The convolutions of the cortex were slightly flattened, and there was a soft cystic mass in the right frontal lobe. Dissection of the frontal lobes revealed the following picture:

Right Frontal Lobe: In the anterior medial portion of the right frontal lobe was a large soft area which measured 3 by .3 by 5 cm. This extended from the

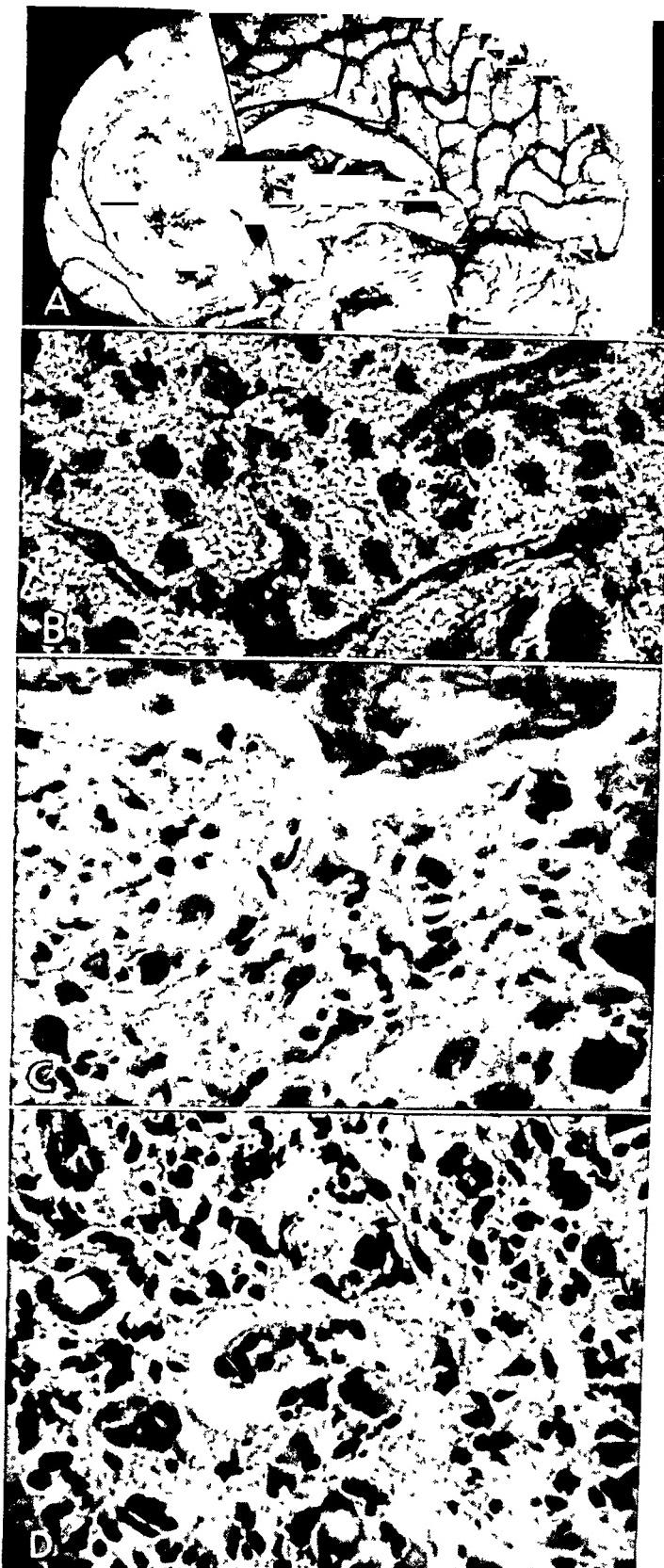


Fig. 3 (case 2).—*A*, invasive tumor of the right frontal lobe reaching the lateral ventricle and a small area of the meninges; *B*, spongioblasts (gold chloride); *C*, thickened vascular walls, hyperchromatic multinucleated cells, mitosis, spongioblasts and neuro-ectodermal cells (hematoxylin and eosin); *D*, same picture with phosphotungstic acid and hematoxylin stain. Reduced from  $\times 450$ .

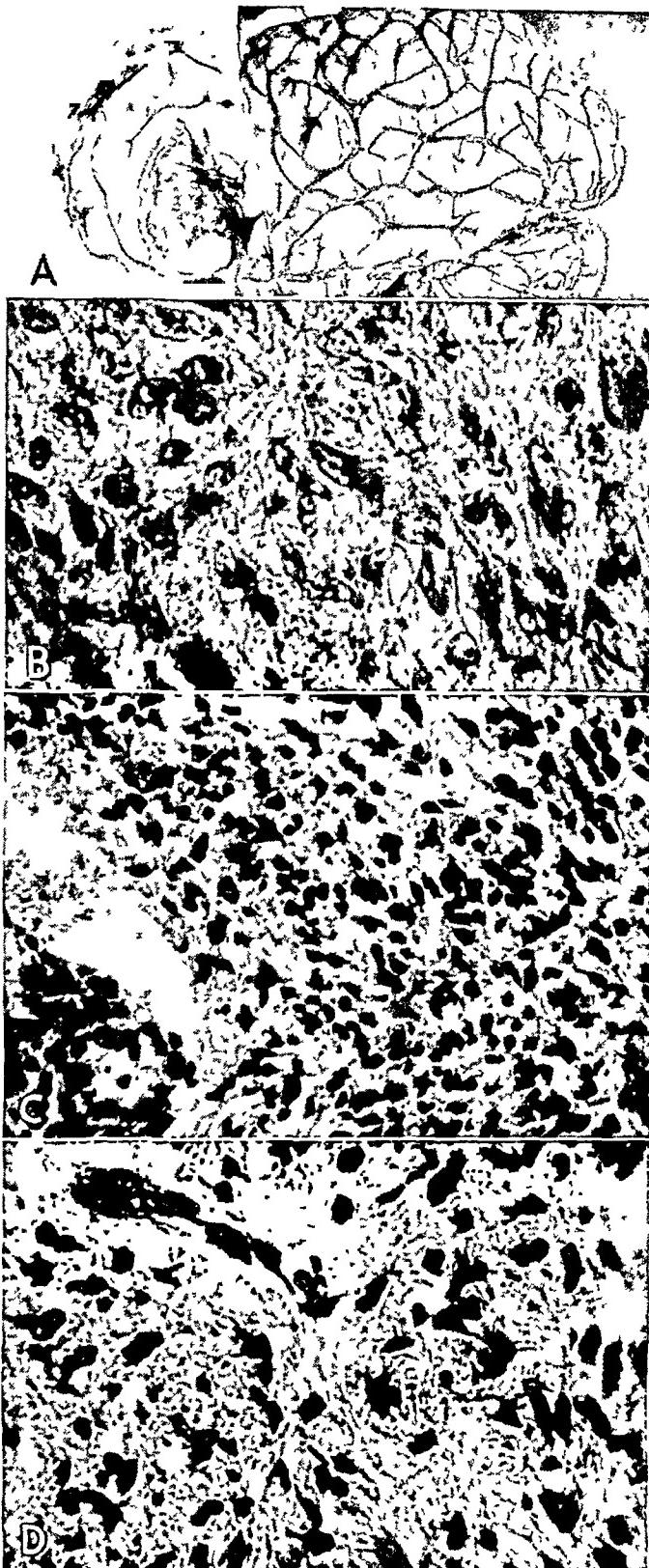


Fig. 4 (case 2)—*A*, tumor of the left frontal lobe; *B*, polar spongioblasts and astrocytes showing glia fibrils (phosphotungstic acid and hematoxylin); *C* and *D*, astrocytes, spongioblasts and neuro-ectodermal cells (hematoxylin and eosin). Reduced from  $\times 450$  and  $\times 300$ .

medial surface of the frontal cortex laterally to surround and encroach on the anterior horn of the lateral ventricle. The entire tumor lay beneath the cortex except where it came to the surface at one point on the medial surface of the lobe. The tumor itself was soft and necrotic. It contained numerous large and small hemorrhages. Areas of necrosis were present which contained many small yellowish-gray particles. The mass appeared quite invasive and showed no tendency to encapsulation. It was considerably less circumscribed than the tumor on the opposite side but was definitely confined to the right frontal lobe.

On microscopic examination the sections presented a "typical" glioblastoma multiforme with great variations in the size and shape of its constituent cells. Many hyperchromatic multinucleated giant cells were scattered throughout the tissue. Mitotic figures were numerous (from one to two per high power field). Many of these were quite bizarre. Apolar spongioblasts and neuro-ectodermal cells constituted the majority of the cells present. There were many variations in the size and shape of these elements, and they were scattered irregularly without any architectural pattern. The presence of astrocytes was not a prominent feature except in a few less cellular areas where they were quite numerous. In those areas the tissue suggested an astrocytoma. Occasional areas of necrosis and free hemorrhage were noted. The tumor tissue was quite vascular. Many of the smaller blood vessels showed both endothelial and adventitial thickening causing partial occlusion of their lumens.

The diagnosis was glioblastoma multiforme of the right frontal lobe.

Left Frontal Lobe (fig. 4) : In the region of the inferior frontal sulcus was a hard firm tumor mass which measured 1.5 by 3 by 4 cm. It barely reached the lateral cortex and did not extend medially as far as the lateral ventricle. The tumor tissue was grayish yellow and rather uniform except for some necrosis about its margins. In some areas the tumor had a tendency to follow along the sulci rather than to destroy all the tissue to the same degree. Little hemorrhage or necrosis was present. There was no encapsulation, and the mass was not sharply circumscribed. No connection could be demonstrated between this tumor and the one in the opposite frontal lobe.

Microscopic examination showed polar spongioblasts predominating in this tissue. These formed wide interlacing bundles. Astrocytes were numerous and formed large fields of tissue which were identical with those of an astrocytoma. In other areas astrocytes were comparatively rare, and many neuro-ectodermal cells were found. Multinucleated giant cells and mitotic figures were not frequent. A few large and small areas of necrosis and hemorrhage were noted. Some of these were fringed by a pseudopalisading of spongioblasts and neuro-ectodermal cells. The tumor was quite vascular, and many of the walls of its blood vessels were greatly thickened. This was the result of endothelial and adventitial overgrowth. In some instances the lumen of the vessel was practically obliterated.

The diagnosis was glioblastoma multiforme of the left frontal lobe.

*Comment.*—This case is considered one in which a glioblastoma multiforme appeared independently in the right and left frontal lobes. A careful study of sections revealed no continuity between the two growths. There was no other evidence of metastasis. However, the possibility of one growth appearing as a metastasis from the other cannot be ruled out entirely since both tumors extended to the meninges. The part of the left tumor enclosed by an area of pia-arachnoid measured only a few millimeters in diameter.

The fibrillary elements of the tumor of the left lobe were more prominent than those of the opposite side, and in many large areas they had the appearance of an astrocytoma.

#### SUMMARY

Two cases of bilateral glioblastoma multiforme are reported.

In case 1 tumors of the left temporal and right parietal lobes were found joined by a filamentous strand of tumor tissue. This passed from one hemisphere to the other just posterior to the splenium of the corpus callosum. It was thought that the left temporal tumor was primary and produced a similar lesion in the right parietal cortex by direct extension into the opposite hemisphere.

In case 2 the tumor was believed to be a bilateral glioblastoma multiforme of the frontal lobe of multicentric origin. No communication or extensions could be demonstrated. It is freely admitted that the possibility of metastasis from one frontal lobe to the other cannot be entirely ruled out since the pia-arachnoid bounded a small portion of each growth. There was, however, no other suggestion of metastasis in the brain.

# CHANGES IN THE BRAIN IN PERNICIOUS ANEMIA

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Changes in the brain associated with pernicious anemia are uncommon, in contrast to those in the spinal cord, which have been well studied. Only a few cases have been reported in which the changes in the brain corresponded histologically with those in subacute combined degeneration of the cord (Preobrazhenski,<sup>1</sup> Lurie,<sup>2</sup> Woltman,<sup>3</sup> Weimann<sup>4</sup> and others).

Lurie studied four cases of pernicious anemia, all of which showed mental symptoms long before death. In three he found in the brain foci of Lichtheim's type and of Preobrazhenski's miliary foci, aside from typical lesions in the spinal cord.

Woltman, from his study of seven cases of pernicious anemia with changes in the cord and brain, came to the conclusion that lesions in the brain run fairly parallel to those of the cord and occur with about the same frequency. Only one patient exhibited mental symptoms, which, however, predominated in Weimann's cases; in one patient they lasted four years and terminated in death.

Both Lurie and Weimann claimed that in the center of the degenerated areas they invariably found a blood vessel, while, according to Woltman, the foci "may or may not be connected with blood vessels."

Of seven cases with a typical picture of subacute combined degeneration of the cord in which the brain was available for study, only one revealed equally typical changes in the brain. Two showed minor changes, such as swollen oligodendroglia, rarefactions, numerous astrocytes and scattered vacuolated areas of minute size. Most of the last-mentioned areas were devoid of cell contents; some harbored glia nuclei. As these changes are often present in other pathologic conditions of the brain, they were not considered significant. Four cases showed no changes in the brain, nor did the patients present mental or cerebral symptoms, except terminal mental disturbances (confusion, hallucinations and stupor).

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From the Division of Neuropathology of the University of Illinois College of Medicine; Dr. G. B. Hassin, Director.

1. Preobrazhenski, P. A.: *Neurol. Centralbl.* **21**:727, 1902.

2. Lurie, L. A.: *Arch. Neurol. & Psychiat.* **2**:67, 1919.

3. Woltman, H. W.: *Arch. Int. Med.* **21**:791, 1918.

4. Weimann, W.: *Ztschr. f. d. ges. Neurol. u. Psychiat.* **92**:433, 1924.

Of these seven cases one is reported, with a description of the changes in the brain; the changes in the spinal cord in this case have been thoroughly discussed by Hassin,<sup>5</sup> who purposely omitted the description of the cerebral lesions, as they were considered striking enough to warrant a separate study.

#### REPORT OF CASE

*History.*—The patient, aged 45, was admitted to the neurologic service of the Cook County Hospital because of difficulty in walking, cramps in the big toes, tingling, reeling gait and "electric shocks" running down his spine. The duration of the illness had been two years.

*Examination and Course.*—He had a spastic gait, an exaggerated patellar reflex, absence of the Achilles reflex and positive Babinski, Rossolimo, Chaddock and similar signs. The cranial nerves, pupils and fundi were normal. The position sense was lost, and the vibratory sense was impaired in the lower extremities.

The number of red blood cells was greatly diminished; the hemoglobin ranged from 65 to 80 per cent, and the color index was 1.2. Differential white and red blood cell counts showed no abnormal forms. Analysis of the stomach revealed achylia gastrica.

The patient grew progressively worse. Atrophy of the right leg and anesthesia below the spine became more evident; incontinence of the bladder set in, and paraplegia in flexion developed. A few days before death from bronchopneumonia he became stuporous and drowsy and was hard to arouse.

The spinal cord presented a typical picture of subacute combined degeneration. The brain revealed no changes macroscopically.

*Microscopic Changes.*—Numerous and extensive foci of degeneration (*status spongiosus*) were scattered in the motor area (fig. 1). On closer examination these areas were found to consist of numerous gliogenous formations (fig. 2), some of which contained basophilic metachromatic granules; others were myelophages or gitter cells  $\alpha$ , but the majority were the classic gitter cells  $\beta$ . Aside from the myelophages and the various gitter cells, numerous cytoplasmic astrocytes were present, which were especially massed around the blood vessels where they formed a wall along the pia glial membrane. The areas of the adjacent spaces of the adventitia were filled with gitter cells, while adventitial elements frequently harbored droplets of fat. The foci were mostly devoid of blood vessels, and only one focus had a blood vessel running through its center. The ganglion cells were somewhat swollen; some exhibited chromatolysis and neuronophagia, but in general the changes in the gray matter were much less marked than in the white matter.

The Bielschowsky and Alzheimer-Mann staining methods revealed in the affected areas, but somewhat remote from the actual foci of softening, rows of vacuoles that contained fragments of swollen axons and myelin, Marchi globules and few myeloclasts. Some of the vacuoles were encircled by a ring emanating from the glia nucleus. In these areas myelophages could be discerned, that is, nerve degeneration was here more advanced, but was fully developed in the areas of *status spongiosus* just described. Careful study of other cortical areas, of the corpus striatum and the optic thalamus revealed no changes. Few vacuoles of minute, miliary size could be seen in the pons.

5. Hassin, G. B.: Arch. Neurol. & Psychiat. 29:855, 1933.



Fig. 1.—Large focus of degeneration in the subcortex of the frontal lobe (status spongiosus); the cortex itself is not involved; Alzheimer-Mann stain.

In the cerebellum the molecular layer and the granular cell layer showed fissures in some sections. These were near the pia mater and contained round, homogeneous, pale bodies (amyloid bodies), some of which were enveloped by delicate fibers of glia. In other places the fissures contained red and other blood cells. Aside from the fissures, there were reactive phenomena in the surrounding glia tissue in the form of fibrillary and cytoplasmic astrocytes. The Purkinje cells, as well as the processes, were tumefied and densely stained.

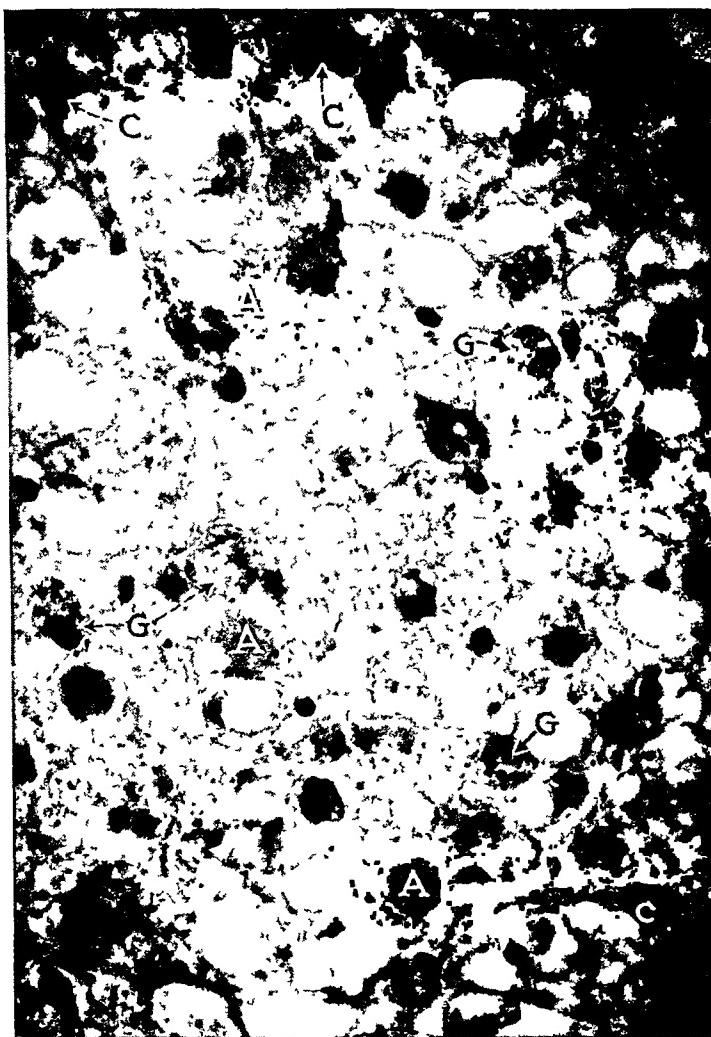


Fig. 2.—Higher power magnification of figure 1; numerous gitter cells (*G*) are interspersed with astrocytes (*C*) and amyloid bodies (*A*); Alzheimer-Mann stain.

The changes in the subarachnoid space were those of so-called aseptic meningitis. The pia-arachnoid was thickened, the meshes contained many small and large lymphocytes, mesothelial cells and numerous gitter cells. In one area there were numerous blue granules, appearing partly as conglomerations and partly within large, nucleated cells. The nature of these could not be ascertained. Probably they were particles of fatlike substances.

The tuft cells of the choroid plexus were swollen; some contained vacuoles, while others were granular. The granules, when stained with toluidine blue, appeared as bluish, dustlike grains of sand scattered over the cytoplasm; other granules appeared as droplets and stained darker. Some such granules, which are often seen in tuft cells of the choroid plexus, stain red with scarlet red and blue with thionine or toluidine blue. They are most likely of a lipoid nature, but in this case their chemical properties could not be studied in detail, as it was not possible to obtain frozen sections.

#### COMMENT

Definite changes were present in the motor areas of the brain. They were of a degenerative nature and of the same histologic structure as similar foci in the spinal cord. As in the spinal cord, only the white matter was affected, though it was less disintegrated. Thus, fewer gitter cells and fewer myelophages were in evidence than are usually seen in the spinal cord under similar conditions. Instead, a number of cytoplasmic glia cells (cytoplasmic astrocytes) were present. Some authors (Henneberg,<sup>6</sup> and Weil and Davison<sup>7</sup>) believe that the widespread destruction of the spinal cord that results in *status spongiosus* is a manifestation of deficiency of glia, which, in their opinion, in pernicious anemia is affected by the same toxic substances that destroy the spinal parenchyma. The presence of cytoplasmic glia in some areas in my case denotes, in contrast, a progressive reaction on the part of the glia, which is always present when the nerve parenchyma undergoes degeneration. The large masses of degenerative products cannot be removed in time by the glia, as the breaking up of the nerve fibers is massive and continuous; hence the vast accumulations of the products, which are typical of early nerve degeneration, occur. In the brain such large accumulations of broken-up and degenerative nerve tissues do not occur: For this reason the repair, in the form of glial reaction, is more manifest. The presence of many scattered cytoplasmic glia cells also denotes a glial reaction against destruction of isolated nerve fibers or their small bundles. Such facts, that is, a manifest reaction of the glia, do not sustain the opinion of the authors cited, but prove that the glia behaves as in any other degenerative condition of the nerves.

Study of the changes in the cortex likewise showed the absence of definite relationship between the blood supply and the degenerative foci; in only one focus was a blood vessel present, but it had no structural changes.

The condition of the subarachnoid space and the choroid plexus deserves consideration. Both showed the presence of lipoids. In the subarachnoid space these were enclosed within the gitter cells; in the choroid plexus they appeared as small granules over the tuft cells. In

6. Henneberg, R.: *Klin. Wchnschr.* **3**:970, 1924.

7. Weil, A., and Davison, C.: *Arch. Neurol. & Psychiat.* **22**:966, 1929.

both instances the lipoids most likely came from the degenerated foci of the cerebral parenchyma, which discharges its waste into the subarachnoid space and the ventricles (Hassin), provoking in them the reactive phenomena already described.

The structural changes did not correspond with the clinical symptoms, for the patient had no cerebral manifestations. The reason is most likely the scarcity of the changes in the brain, which appeared only as small foci. As has been pointed out, they did not affect the cortex, but were situated in the subcortical area of the motor region. Cerebral manifestations that might have been produced by these minute subcortical foci may have been overshadowed by the signs of degeneration of the spinal cord.

# EXTENSIVE GENERALIZED TORULOSIS IN A CHETAH OR HUNTING LEOPARD (*CYNAELURUS JUBATUS*)

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AND

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Torulosis, an infection caused by *Torula histolytica*, is known to occur in man throughout the world.<sup>1</sup> Its chief characteristic is the tendency for the yeast cells to localize in the central nervous system, where they produce chronic cystic meningo-encephalitis. The symptoms are mainly those of meningitis and are referable to the lesions in the brain, cord and meninges. Involvement of other organs is uncommon.

The histopathology of torulosis of the central nervous system is peculiar and characteristic. The irregular thickening of the meninges by fibrosis, together with infiltration by mononuclear phagocytes, small giant cells, lymphocytes and plasma cells is not particularly distinctive. However, polymorphonuclear leukocytes are conspicuous by their absence. Highly characteristic, on the other hand, are the large number of yeast cells and the type of focal lesion which they cause. The cells are contained in great numbers within the cytoplasm of the mononuclears and giant cells, but others are free in the tissue spaces. When lesions are present in the brain they may be either cystic or solid, single or multilocular. Singularly, inflammatory reaction is lacking when the cysts occur in the brain substance, but in the meninges there are notable accumulations of mononuclears and infiltrations of perivascular tissues by lymphocytes and plasma cells.

In other organs the histopathologic picture may be essentially the same. At places there is no recognizable tissue reaction against the micro-organisms, while at other places the lesions may consist of areas of granulomatous change in which normal tissues are replaced by colonies of yeast cells surrounded by (usually minor) accumulations of mononuclear and giant cells. Again, a reaction like that of tuberculosis, with its zones of endothelioid cells and lymphocytes, may occur around the parasites, but this is relatively uncommon.

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From the Laboratory of Comparative Pathology, Philadelphia Zoological Gardens, and the Laboratory of Dermatological Research, University of Pennsylvania.

1. Stoddard, J. L., and Cutler, E. C.: Monograph Rockefeller Institute no. 6, 1916. Freeman, W.: J. f. Psychol. u. Neurol. **43**:236, 1931.

So far as can be determined from the available literature, only one other case of spontaneous torulosis has been reported in a lower animal; this occurred in a horse, in which the disease was confined to the lungs.<sup>2</sup> The disease has not been reported in the large collection of wild animals maintained by the London Zoological Society,<sup>3</sup> where the beasts are closely examined at necropsy, or from other menageries. Furthermore, generalization such as was observed in this animal occurred in only three of the forty-four cases in man recently collected by Freeman.<sup>4</sup> Indeed, the massiveness of involvement, particularly of the kidneys and spleen, has not been matched in any case thus far reported, either in man or in lower animals.

#### REPORT OF CASE

*History.*—The present case, the first instance of torulosis encountered in more than ten thousand consecutive autopsies performed as a routine on wild animals dying in the Philadelphia Zoological Gardens, concerned a male chetah or hunting leopard, *Cynaelurus jubatus*. This animal, which had been acquired as one of two cubs, had grown with its fellow to maturity while on exhibition in the garden for five and one-half years. Symptoms were not noted until about a month before death, when the animal was reported as "stiff in the hindlegs." One week later he refused all food except milk and appeared to be completely blind. Death occurred five days later.

*Necropsy.*—At necropsy the body was fairly well nourished. The skin and subcutaneous tissue, as well as the skeleton and musculature, were normal.

The spleen was approximately four times normal size, measuring 38 by 12 by 7 cm. in its greatest dimensions; it was firm and extremely friable. The surface was irregularly elevated by large numbers of closely placed and occasionally confluent, firm, roughly spherical, pale gray nodular masses, several of which were pedunculated. They varied in diameter from 0.1 to 5 cm., and had indistinct outlines which merged with surrounding tissue. On section, the organ was found to be involved throughout its entire extent, the tissue being pale yellow in general, with red-brown and yellowish mottlings. It was firm and friable but free from gross evidence of softening or degeneration. Hemorrhagic areas were numerous on the surface of the section, especially about the larger nodules.

The kidneys were about twice normal size, measuring 10 by 6 by 4 cm. The capsules were irregularly adherent. The external surfaces were largely covered by numbers of small, firm, pale gray, ovoid nodules with indistinct outlines, measuring about 0.5 cm. on cross-section. On the surface of the section similar nodules were found, particularly along the corticomedullary junction, where they sometimes formed continuous, irregular, bulging zones much paler than the surrounding tissue, but apparently of the same consistency.

The retroperitoneal lymph nodes were enlarged, firm and pale gray; the normal architecture was not recognizable grossly.

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2. Frothingham, L.: J. M. Research **3**:31, 1902.

3. Proc. London Zool. Soc., 1900-1932.

4. Fitchett and Weidman have reported a fourth case, with involvement of the kidneys, pancreas, spleen, and lymph nodes, in addition to the cerebrospinal nervous system (Arch. Path. **18**:225, 1934).

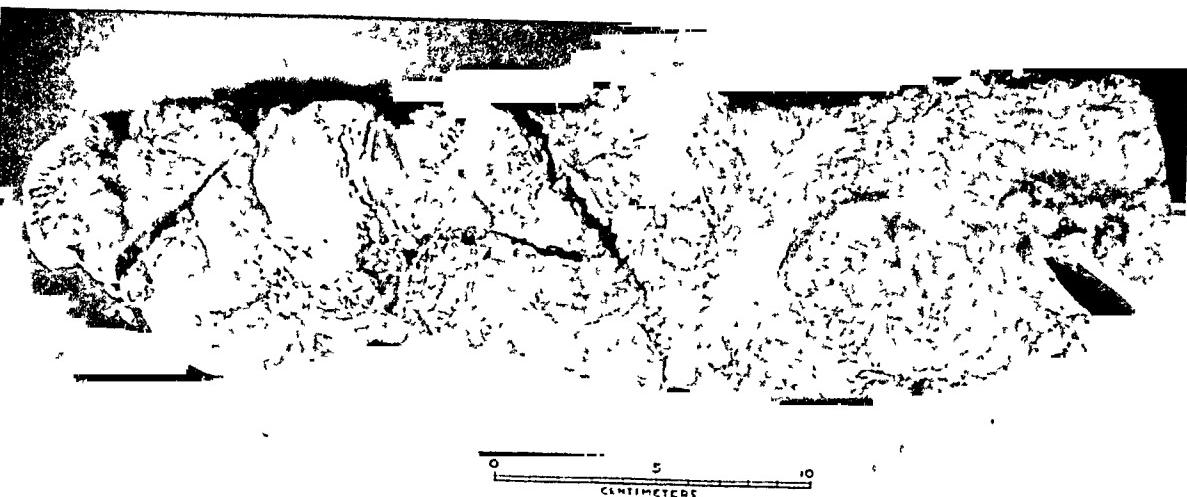


Fig. 1.—Hugely enlarged spleen covered by white nodules of torular infiltration.



Fig. 2.—Kidney; this is like the spleen in its huge enlargement and in the presence of white nodules.

The meninges did not appear thickened or adherent when the brain was first removed. Furthermore, there was no evidence of parenchymatous involvement until the brain was examined after fixation. This is also frequently the case in man. After fixation, numbers of small, roughly spherical areas of softening from 0.1 to 0.2 mm. in diameter were found in the cerebral cortex, basal ganglia and pons. As seen on the cut surface of the brain, these areas appeared as sharply outlined individual cysts filled with homogeneous, gray-white, translucent jelly-like substance. The latter was retracted at the center, giving a shallow depression. Inflammatory response was lacking around the lesions.

The spinal cord was not examined.

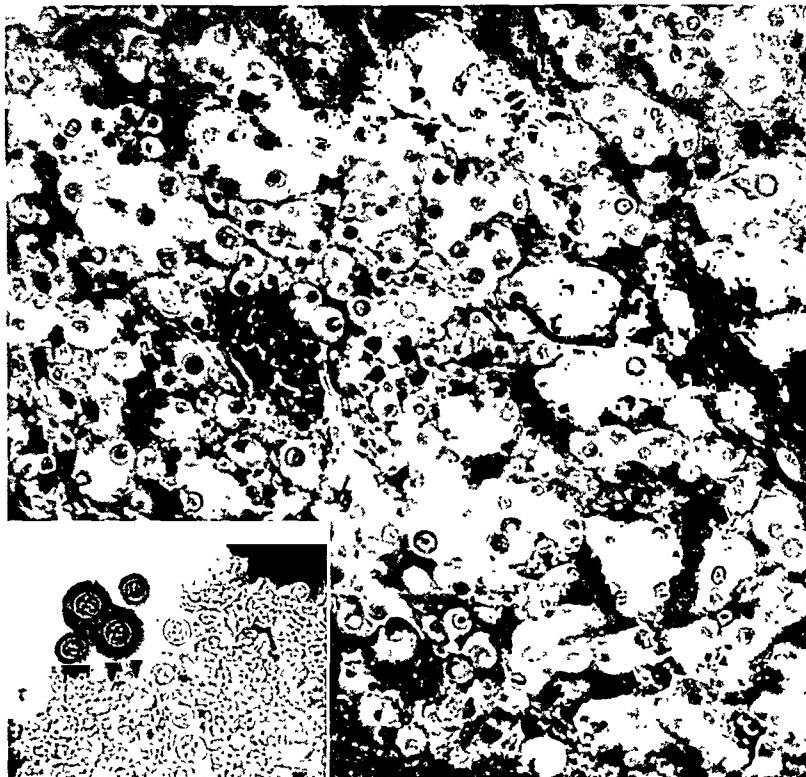


Fig. 3.—Diffuse infiltration of the splenic pulp by yeast cells. The insert shows a group of torula cells scraped from the surface of the spleen, with their characteristic halos (mucinoid envelops); wet india ink preparation.

*Microscopic Notes.*—Spleen: Except for the capsule, some fragments of trabeculae and an occasional splenic nodule, nothing remained of the normal structures. The tissue was replaced largely by an eosinophilic, hyaline reticulum containing occasional fibroblasts and indistinctly outlined giant cells. Contained in this reticulum were numerous small (from 4 to 10 microns), doubly contoured spherical bodies, which will be described in detail in a later paragraph. They occurred singly or in groups in the reticulum and in the giant cells.

In the interstices of the reticulum were also occasional multinucleated cells, together with macrophages containing the invading organism, not to mention the large numbers of unengulfed organisms. Occasional areas in the trabeculae and capsule had the appearance of recent involvement, as shown by the presence of

a young type of fibrous tissue and small numbers of macrophages. Other than this, inflammatory reaction was entirely lacking.

Kidneys: The parenchyma of both the cortex and the medulla was extensively and irregularly replaced by granulation tissue. Macrophages and small giant cells were numerous throughout the lesions, almost all of which were distended by large and small yeast cells. The nodules on the capsular surface were also made up of such inflammatory growths and infiltrations. Unengulfed yeast cells were rare in the tissue spaces. The most conspicuous of such "free" groups were located within the lumens of glomerular capillaries or in tubules. In the latter situation the surrounding tissues appeared to have undergone lysis, and macro-

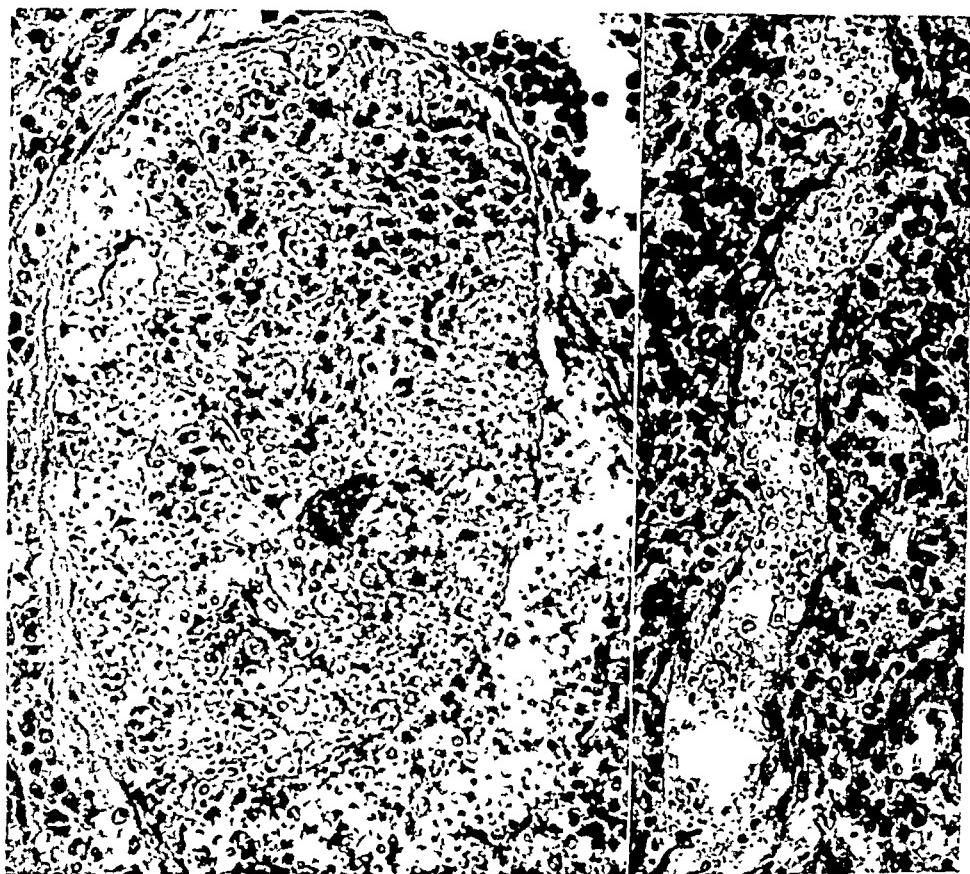


Fig. 4.—Section of the kidney; details of the yeast cells do not show up well, but there are great numbers in both the tuft itself and the subcapsular space; the tubular lumens also contain yeast cells.

phages were absent. Yeast cells were also occasionally observed filling the lumens of small blood vessels. Plasma cells and lymphocytes were present in dense masses, both adjacent to these lesions and apart from them.

Lymph Nodes: The retroperitoneal lymph nodes were practically completely necrotic, except for the capsule and for the occasional persistence of blood vessels. The latter were surrounded by a narrow band of lymphocytes. Yeast cells lying either free or in an eosinophilic coagulum replaced the lymphoid elements.

In the mesenteric lymph nodes the yeast cells were arranged in the form of small colonies. These were located mainly in the cortical nodes which consisted of more or less discrete foci in which the normal lymphoid elements were

replaced by macrophages containing numbers of parasites. The medullary and peripheral sinuses of the lymph nodes were often distended by a similar type of macrophage. Few of these contained yeast cells.

**Liver:** Sections of this organ contained numerous small (from 50 to 200 microns) sharply outlined rounded areas in which the parenchyma had been replaced by accumulations of macrophages and small giant cells. The yeast cells lay within the cytoplasms of the latter. Occasionally, polymorphonuclear neutrophils and lymphocytes were also present within the smaller lesions. These foci were not related to any special structures of the liver (portal vein, artery or duct).

**Suprarenal Glands:** Lesions similar in most respects to those of the liver were present in the midzone of the cortex of the suprarenal gland. In larger lesions the inflammatory elements had undergone necrosis in the central parts, leaving the yeast cells free in a turbid basophilic mass.

**Lungs:** Changes in these organs which may be attributed to torulosis were of minute size and consisted of circular or fusiform thickenings of the alveolar walls or interlobular septums. There was a moderate fibrous tissue overgrowth in these areas, together with accumulations of macrophages (containing parasites) and lymphocytes.

**Brain:** There was an irregular thickening of the pia-arachnoid, which was most pronounced in the depths of sulci. This was largely referable to hyperplasia of fibrous tissue, edema and cellular infiltrations and was most marked when yeast cells were also present. Many of the latter were contained within the cytoplasm of macrophages, but they also occurred free in tissue spaces. Occasionally, groups of parasites lay in spaces in the thickened meninges. Within the parenchymatous nerve tissue the lesions due to torulosis consisted of single or multilocular cysts. The smaller of these were made up mainly of masses of closely placed mononuclear phagocytes and small giant cells, which had replaced almost all the normal tissue elements. These cells contained large numbers of the parasites, but the latter also occurred occasionally between the cells. No inflammatory response was evident around these lesions, except for conspicuous accumulations of lymphocytes about the blood vessels which passed adjacently to the periphery or into the cysts. In many cysts none of the normal cerebral tissue remained except patent blood vessels—arterioles with dense mantles of lymphocytes and widely distended perivascular lymph spaces. In other cysts, bands of glial tissue, usually associated with blood vessels, divided the lesions. Peripheral compression of tissues about the cysts was inconspicuous. Macrophages containing yeast cells were present in the nerve tissue immediately adjacent to the lesion.

#### THE INFECTING ORGANISM

Unfortunately, an attempt was not made to cultivate the organism from tissues at necropsy. In fact, infection was not even suspected at that time, since appearances seemed to indicate a neoplasm. However, examination of the sections and wet smears from the various organs allowed identification of the micro-organism within certain limits, particularly since its morphologic appearance and singular tissue changes corresponded so closely with those already well known in man.

In sections, the organism was sometimes perfectly spherical, but usually was distorted. It varied between 4 and 12 microns in diameter. It had a thick, refractile shell surrounded by a relatively thick capsule (this was best seen in the containing one or two large clear vacuoles and a small amount of chromatoid larger cells). In the interior there was a small amount of eosinophilic cytoplasm

substance. As a rule, the cells free in tissue spaces lay in a thin, eosin-staining matrix. In the case of larger cells, one or several small ones frequently occurred in juxtaposition. This was clearly an expression of budding, since all stages, from the first extension of the protoplasm to the formation of the daughter cell (or even cells equal in size to the parent cell), were seen. Reproduction within large cells (asci) was not recognized.

In wet smears from the involved organs, prepared by scraping material from the cut surface and suspending it in saline solution or india ink, the bodies appeared as highly refractile spheres from 4 to 14 microns in diameter. Many showed budding; this was sometimes multiple, and occasionally the buds were arranged in a chain consisting of two or three small daughter cells. In such preparations the thick mucinoid capsule was especially evident, and within it, in turn, the double capsule around the parasite could be seen to advantage.

There can be little doubt that the infecting organism in this case was a yeast, but it is hardly possible to make further and final identification without cultures. Still, the limited number of species involved in torular infection, their tendency to be confined to certain anatomic locations—notably the cerebrospinal system—the uniformity with which *T. histolytica* is identified in cases of cerebrospinal involvement and the singular type of tissue reaction associated with such infection must permit tentative identification of the parasite in this case as *T. histolytica*.

#### ANIMAL TESTS

These tests centered around the question whether Felidae in general were susceptible to infection with *T. histolytica*. We found that rats are highly susceptible on intrameningeal inoculation, and Freeman's monograph indicates the consistency with which guinea-pigs also can be infected. To our surprise, the cat was found to be highly resistant, even on intrameningeal inoculation. In each animal the brain (and sometimes other organs) was examined histologically.

CAT 1.—This animal was inoculated with strain 664, which had produced frank torular meningitis which killed a rat within seven weeks. The cat died within ten days, without any evidence of successful invasion.

CAT 2.—This animal was inoculated in the same manner as cat 1. It was killed at the end of three months. The meninges were moderately infiltrated with yeast cells.

CAT 3.—Strain 1725, employed here, was the most virulent of twenty strains of *T. histolytica* with which we had worked. It had induced generalized torulosis in each of four rats tested, involving no less than three, and sometimes five, of the following organs: brain, lungs, liver, spleen and kidney. The cat was killed at the end of one month, but did not disclose any lesions either at necropsy or on histologic examination. *T. histolytica* was, however, secured from the meninges by retroculture.

CAT 4.—Strain 2249 was one recently cultured from a human patient with a widely generalized infection. When injected into a rat, it involved the brain, pancreas, kidney and spleen. The cat died within four and one-half weeks, without any evidence of torular infection.

CAT 5.—This animal was inoculated with the same strain employed in cat 4. It died within nine days, showing torular infiltration of the cerebral meninges of such a mild grade that it suggested an infection that was dying out.

The consistency with which all five animals were unaffected, even though three different strains (two of which were highly infectious for rats) were

employed, in the face of such a severe test as injections directly into the meninges, stamps the cat as among the genera resistant to torulosis. From this it might be (superficially) deduced, by analogy, that the chetah, a co-feline is similarly resistant to *T. histolytica* and that a different species of *Torula* was concerned in it. We do not subscribe to such a conclusion. It is well known that among rodents the guinea-pig is relatively resistant to experimental blastomycosis, whereas the rat is highly susceptible. Indeed, until it is proved otherwise, we believe that the burden of proof lies on those who would discredit *T. histolytica* as the cause of the morbid anatomic changes which were so distinctive in the chetah and which compared so closely in morphologic features and tissue reactions with torulosis in man.

#### SUMMARY

A second instance of torular infection in a lower animal is recorded in a chetah which died in captivity. It is only the fourth instance in which the infection was widely generalized, either in man or in lower animals. The spleen and kidneys were massively enlarged, far beyond the size yet recorded for man in those organs. Lesions in the cerebral cortex were cystic, as they frequently are in man. Although the organism was not secured in culture, its morphologic features in lesions and the distinctive tissue changes were so similar to those seen in man that the organism can be identified tentatively, at least, as *T. histolytica*. Cats were found to be strongly resistant to infection with three different strains of rat-virulent *T. histolytica* on intrameningeal inoculation.

# INFLUENCE OF DRUGS USED IN ANTISYPHILITIC THERAPY ON THE RETICULO-ENDOTHELIAL SYSTEM

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Little is definitely known concerning the manner of the prophylactic or curative action of the drugs used in the treatment of syphilis. Since Ehrlich applied his side-chain theory of immunity to chemotherapy many investigators have advanced different theories as to the mode of action of arsenicals *in vivo*. There is as yet a lack of complete agreement, but the generally accepted idea is that the action is probably due to a combination of factors rather than to one factor alone. Kolmer<sup>1</sup> suggested that a new substance may be formed by combination between the injected drug and the proteins of the blood or tissue; there may be chemical change by oxidation or reduction, or antibody formation may be stimulated by the injected drugs. The fact that arsenical drugs, such as the arsphenamines, do not exert the same spirocheticidal action *in vitro* that they do *in vivo* shows that the action is not a simple chemical one of the drug on the parasite.

Voegtlin and Smith<sup>2</sup> first advanced the hypothesis that arsenical compounds must be changed to one type, namely, the trivalent oxides RAs:O, before exerting their principal toxic action. These substances are formed by oxidation of trivalent and reduction of pentavalent arsenicals, respectively. Subsequent work by Voegtlin and others supported this idea, and Rosenthal<sup>3a</sup> devised a method of demonstrating arsine oxide *in vitro* and *in vivo*. He was able to recover from 10 to 12 per cent of injected arsphenamine as arsine oxide from the liver of the rat from three to four hours after injection. A comparable concentration was demonstrated in the rat kidney after the injection of neo-

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This work was aided by funds from the Committee on Research in Syphilis, Incorporated, the Douglas Smith Foundation and the A. B. Kuppenheimer Foundation.

1. Kolmer, J. A.: Principles and Practice of Chemotherapy, with Special Reference to the Specific and General Treatment of Syphilis, Philadelphia, W. B. Saunders Company, 1926.

2. Voegtlin, Carl, and Smith, H. W.: J. Pharmacol. & Exper. Therap. **15**: 475, 1920.

3. Rosenthal, S. M.: (a) Pub. Health Rep. **47**:933, 1932; (b) *ibid.* **47**:241, 1932.

arsphenamine. Failure to demonstrate arsine oxide in other organs and tissues was attributed to probable low concentrations.

Some interesting work has been done in an endeavor to explain the mechanism of the oxidation and reduction reactions in vivo. Hopkins and Dixon<sup>4</sup> showed that glutathione is concerned in biologic oxidation and reduction. Hopkins<sup>5</sup> isolated from mammalian muscle a tripeptide containing cysteine and showed that the sulphhydryl group in that molecule can undergo reversible oxidation-reduction. Voegtlin, Dyer and Leonard<sup>6</sup> explained the toxicity of the arsine oxides by the hypothesis that they are specific poisons for the sulphhydryl group in glutathione and possible other sulphhydryl compounds which may occur in protoplasm. Rosenthal<sup>3b</sup> demonstrated the ability of sulphhydryl groups to combine with arsenoxide, but not with pentavalent arsenic. He also showed that trivalent arsenic does not combine with native proteins containing no sulphhydryl groups.

Aschoff<sup>7</sup> revived interest in fixed phagocytic cells of the body by grouping them together under the name of the reticulo-endothelial system. This includes the phagocytic cells of the spleen, liver, bone marrow and lymph nodes and the histiocytes of the connective tissue. This system can be readily studied because the cells phagocytose and retain for varying periods such substances as lithium carmine, trypan blue, india ink, saccharated iron oxide and colloidal metallic compounds, e. g., of silver, mercury, bismuth and copper. When the reticulo-endothelial system has taken up large quantities of these substances it is said to be "blocked," with reduction of certain functions. It has been shown, however, that complete dysfunction does not obtain. Various functions have been assigned to this system of cells, including, in addition to their phagocytic properties, participation in antibody formation and alteration of chemical substances injected for therapeutic purposes. Several observations have been made relative to drugs used in syphilitotherapy. Del Baere<sup>8</sup> showed that neoarsphenamine remains longer in the circulation after an injection of solution of pituitary and also that after an injection of neoarsphenamine substances such as congo red and emulsified fat, which are known to leave the blood stream by way of the reticulo-endothelial system, remain longer in the circulation. This is supportive evidence of Schlossberger's<sup>9</sup> idea that neoarsphenamine leaves the blood stream, at least in part, by way of the reticulo-endothelial system. This is probably due to the fact that it is a semi-

4. Hopkins, F. G., and Dixon, M.: J. Biol. Chem. **54**:527, 1922.

5. Hopkins, F. G.: Biochem. J. **15**:286, 1921.

6. Voegtlin, Dyer and Leonard: Pub. Health Rep. **47**:1882, 1932.

7. Aschoff, L.: Lectures on Pathology, New York, Paul B. Hoeber, Inc., 1924.

8. Del Baere, L. J.: Wien. klin. Wchnschr. **38**:1131, 1925.

9. Schlossberger, H.: Centralbl. f. Bakt. (Abt. 1) **110**:210, 1928.

colloid. The therapeutic action of arsphenamine has been shown to be absent in mice and rabbits which have been infected with the organism of relapsing fever and which have been splenectomized and "blocked" with saccharated iron. When this work was repeated in animals infected with *Spirochaeta pallida* and *Trypanosoma equiperdum* the results were otherwise. Schamberg, Kolmer, Rule and Madden<sup>10</sup> came to the following conclusions:

1. Partial "blockade" of the cells of the reticulo-endothelial system of rats by intravenous injections of India ink slightly increased resistance to infection with *Tryp. equiperdum*.
2. This partial "blockade," however, had no demonstrable effects upon the trypanocidal activity of arsphenamine and neoarsphenamine.
3. Partial "blockade" of rabbits by intravenous injections of India ink had no appreciable effects upon infection with *Spirochaeta pallida*.
4. This partial "blockade" of rabbits likewise had no demonstrable effect upon the spirocheticidal properties of arsphenamine and neoarsphenamine.
5. Removal of the spleens of rats had no appreciable effects upon the development and course of infection with *Tryp. equiperdum* inoculated intraperitoneally five to seven days later.
6. Splenectomy, however, definitely reduced the trypanocidal activity of both neoarsphenamine and arsphenamine given five to seven days later, since the minimal curative doses were almost twice as much as observed among control animals.
7. Removal of the spleens of rabbits had no appreciable influence upon the development and course of acute testicular syphilis of rabbits following inoculation with *Spirochaeta pallida* approximately three to four weeks after operation.
8. Splenectomy, however, definitely increased rather than decreased the spirocheticidal activity of arsphenamine.
9. While partial "blockade" of the reticulo-endothelial system with India ink had no appreciable effects upon the trypanocidal and spirocheticidal activity of arsphenamine and neoarsphenamine, it is apparent that splenectomy profoundly influences the chemotherapeutic activity of these compounds.

Golowitzina<sup>11</sup> concluded that the reticulo-endothelial system does not constitute the cell complex on which oxidation or reduction in the organism depends.

Goldzieher and Peck<sup>12</sup> blocked white rats with trypan blue, injected mercuric chloride or sulpharsphenamine, killed the animals at various times thereafter and examined the liver and spleen. They found an increase in the number and size of the Kupffer cells of the liver and a noticeable enlargement and vacuolation of the nuclei in the animals treated with mercuric chloride. The enlargement of the cells decreased after a twenty-four hour period, but the nuclear changes persisted. After forty-eight hours there was no demonstrable difference in the

10. Schamberg, J. F.; Kolmer, J. A.; Rule, A., and Madden, B.: Am. J. Syph. **17**:176, 1933.

11. Golowitzina, K. A.: Ztschr. f. Immunitätsforsch. u. exper. Therap. **71**:541, 1931.

12. Goldzieher, M. A., and Peck, S. M.: Arch. Path. **3**:635, 1927.

microscopic picture between the dyed and the undyed animals. In the rats given injections of sulpharsphenamine there was an enormous swelling of the Kupffer cells two hours after the last injection. The nuclear changes were the same as in the animals treated with mercuric chloride. These authors concluded that there was direct stimulation of these cells by the drugs administered.

Von Jancsó<sup>13</sup> suggested the use of the arsphenamines as vital stains for the reticulo-endothelial system, since they are taken up by these cells and may be demonstrated by a silver method. Arsphenamine, silver arsphenamine and neosilver arsphenamine could be found in the reticulo-endothelial system as long as three weeks after injection, and small emboli in capillaries also gave the reaction. After subcutaneous injection the reaction was positive in histiocytes of the skin and subcutaneous tissue. Neoarsphenamine and sulpharsphenamine were found especially in the connective tissue of abdominal organs and only to a slight extent in reticulo-endothelial cells.

#### EXPERIMENTS

Several series of white rats of approximately the same age and weight were used. Preliminary results were irregular in stock rats, especially in regard to size and vacuolation of the reticulo-endothelial cells. This irregularity of results was also found in control animals. It was thought to be due to previous or present infection, so the work was repeated, using nondiseased rats of the strain obtained from the Wistar Institute. Twenty rats comprised a series; ten were dyed and ten undyed. The dyeing was done by repeated subcutaneous injections of trypan blue.

Five of each group, dyed and undyed, were treated, and the other five were kept as controls. The following drugs were administered intravenously: neo-arsphenamine, thio-arsene (disodium salt of bismuth [p-sulphophenyl] [p-acetamido-phenyl] dithio-arsenite), colloidal bismuth, mercurosal, tryparsamide and gold sodium thiosulphate. The following drugs were administered intramuscularly: sulpharsphenamine, bismuth arsphenamine sulphonate, potassium bismuth tartrate, mercuric chloride and sodium cacodylate. The doses were about seven times the therapeutic doses in man, which are comparable to those used by Goldzieher and Peck.<sup>12</sup> The intramuscular injections were given three times at five day intervals, and the intravenous injections three times at weekly intervals. The animals were killed by bleeding under ether narcosis at intervals of one, three, six, twenty-four and forty-eight hours after the last injection, and the liver and spleen were examined.

Frozen sections were studied by the following methods:

1. Silver method of von Jancsó. Tissue is fixed from one to four days in solution of formaldehyde, and frozen sections are made. To a 1.5 per cent aqueous solution of silver nitrate as many drops of ammonium hydroxide are added as are necessary to make the solution again clear. An equal amount of double distilled purest glycerin is added. Sections are placed in this solution for from thirty to thirty-five minutes. The arsphenamine derivative stains brown or black. The sections are washed in distilled water for one minute and placed in 1 per cent aqueous sodium thiosulphate solution for from three to ten minutes.

13. von Jancsó, N. Jr.: Ztschr. f. d. ges. exper. Med. 61:63, 1928.

They are then dehydrated and mounted in balsam. They may be counterstained if desired. This method depends on the reduction of ammoniacal silver nitrate by arsphenamine in the tissues. As is the case with all silver methods, results were irregular in various sections and even in different parts of the same section in our study. Positive results were found only with the arsphenamines. In the case of thio-arsene, which is a good spirocheticidal drug somewhat different chemically from arsphenamine, no change of any kind was seen in the tissues. Positive results were manifested by the appearance of granules in macrophages (fig. 1) and browning of some of the connective tissue structures as well as, occasionally, the nuclei.

2. Nitroprusside reaction. To determine whether sulphhydryl compounds were present in appreciable amount in the macrophages of the liver and spleen and



Fig. 1.—Liver of rat which had received neoarsphenamine intravenously; the Kupffer cells contain dark brown granules; von Jancsó silver method.

might take part in the oxidation-reduction reactions, they were examined by the following method:<sup>14</sup> Frozen sections about 40 microns thick are floated onto microscopic slides with as little soaking in water as possible. They are then treated successively with 10 per cent zinc chloride, 5 per cent sodium nitroprusside in 15 per cent ammonium sulphate and 2 per cent ammonia. The reagents are added from a dropping pipet, and each is added about thirty seconds after the preceding one. It is desirable to pour off each reagent, without washing, before the next is added. A positive reaction consists of a pinkish coloration in tissues containing large amounts of sulphhydryl derivatives. In the animals studied the reaction was negative, which shows that the substances were not present in sufficient quantity to give a positive reaction, such as can be obtained in sections of epidermis.

14. Percival, G. H., and Stewart, C. P.: Brit. J. Dermat. 42:215, 1930.

Paraffin sections were studied by the following methods:

1. Delafield's hematoxylin stain, as used by Goldzieher and Peck.<sup>12</sup> By this method the general morphology of the tissue could be studied. With stock rats certain changes were noted, namely, enlargement of macrophages, with occasional phagocytosis of red cells. These findings were not uniform and were also seen in controls. When the work was repeated, using an infection-free strain of rats obtained from the Wistar Institute, no appreciable changes in the macrophages could be seen, except enlargement in the case of the animals in which a dye had been injected. Certain granules were seen in the macrophages, and the following method was used for their study.

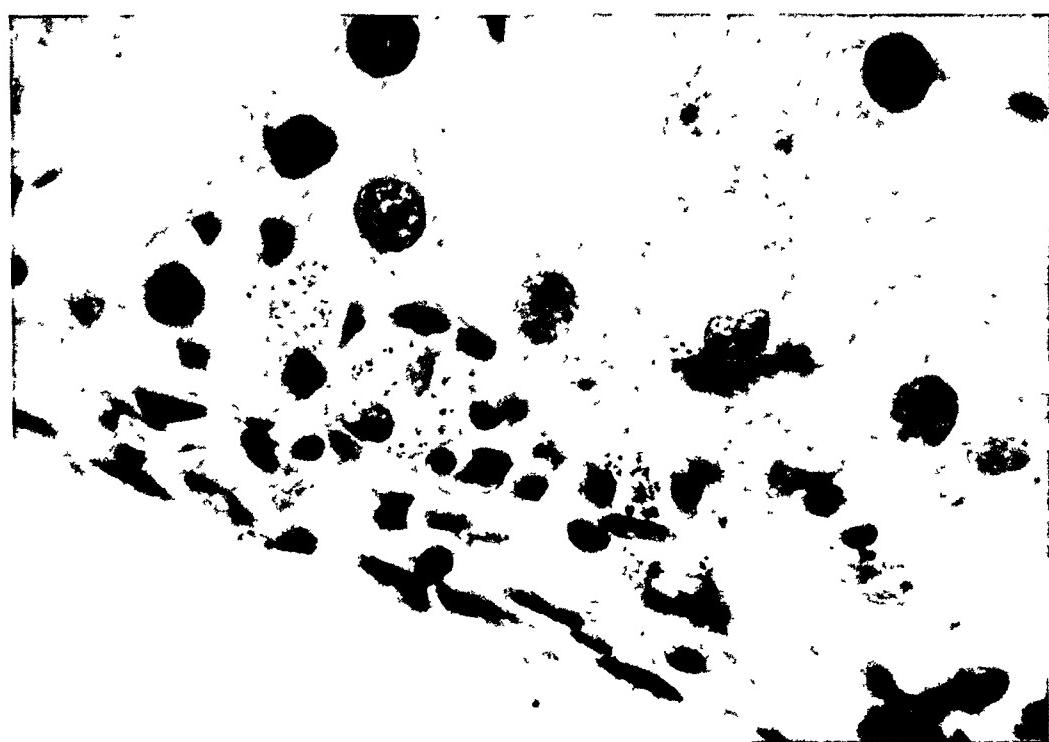


Fig. 2.—Liver of rat which had received neoarsphenamine; brownish granules are seen in macrophages about the central vein of the lobule; Delafield's hematoxylin stain.

2. Pyronine methyl green. This method stained alcohol-fixed tissue sufficiently for orientation without masking cell inclusions. It also furnished a contrast stain to the yellowish-brown granules which were found in the animals studied after the administration of certain drugs. Following the injection of bismarsen, potassium bismuth tartrate, sodium cacodylate and tryparsamide, no granules were seen in the macrophages. Following the injection of neoarsphenamine, brownish granules were seen in Kupffer cells about the central vein of the lobule (fig. 2) one, three and six hours after death and about the large vessels in the spleen three hours after death. Following the injection of sulpharsphenamine, granules were found in the spleen one and six hours after death and in the liver three and six hours after death. After the injection of colloidal bismuth, brown granules were found concentrated in the intermediate zone in all sections of liver and in the spleen

six, twenty-four and forty-eight hours after death. Following the administration of mercuric chloride, brownish granules were found in the liver and spleen six and twenty-four hours after the last treatment. After the injection of gold sodium thiosulphate, granules were found in the liver of the animals examined three and six hours after death and in the spleen of those examined six hours after death. All granules did not give the iron reaction. Similar granules were found in the liver of a patient who died with arsphenamine dermatitis. Professor Pautrier of the University of Strasbourg kindly permitted us to study sections from this patient.

3. Hydrogen sulphide method of Justus, as modified by Brünauer and later by Osborne.<sup>15</sup> The sections prepared by this method did not differ from the control sections, except that they contained some of the brownish granules already mentioned. All sections, including the controls, contained the greenish crystals described by all workers, which were assumed by them to be arsenic trisulphide. Further study of this method by Tannenholz and Muir<sup>16</sup> showed that the visible bodies were crystals. They were not soluble in sodium hydroxide, which will dissolve arsenic trisulphide. Treatment of egg albumin alone and of egg albumin containing sodium arsenite, red blood cells and blood serum, respectively, with hydrogen sulphide produced the same crystals; these were present in greater numbers in the material containing blood serum. An exsanguinated heart showed the crystals only in the muscle cells, while in a heart containing blood they were present throughout. Treatment of the tissue with hydrogen sulphide in an acid medium before fixation did not produce the crystals. Alcoholic fixation before treatment with hydrogen sulphide also was not followed by production of the crystals. It was concluded that the crystals may be a sulphur-protein compound and that they have not yet been proved to be crystals of arsenic trisulphide. In an effort to demonstrate arsenic in tissues, Tannenholz and Muir treated sections with silver nitrate, but could find no difference between those containing arsenic and the controls. The colorimetric method for demonstrating arsenic and phosphorus, which was introduced by Denigès and later modified by Truog and Meyer, was found unsatisfactory, because of large amounts of phosphorus and iron.

It is probably correct to say that at present there is no method that will identify arsenic in microscopic sections.

4. Iron reaction. Kyes utilized the Berlin blue reaction to study macrophages in pigeons. This method was found to be not applicable to human beings, owing to the small amount of hemosiderin contained in the macrophages.

#### COMMENT

When nondiseased rats were used, the marked changes in the cells of the reticulo-endothelial system reported by Goldzieher and Peck were not seen, although similar changes were seen irregularly in stock rats. By means of the silver impregnation method of von Jancsó, granules might be seen in the macrophages in some sections, and in sections stained by pyronine methyl green brownish granules might be seen in macrophages in various portions of the lobules of the liver. There is no doubt that the reticulo-endothelial cells participate in the handling of various antisyphilitic drugs, but whether they take them up as they

15. Osborne, E. D.: Arch. Dermat. & Syph. **12**:773, 1925; **18**:37, 1928.  
Osborne, E. D.; Putnam, E. D., and Hitchcock, B. S.: ibid. **25**:419, 1932.

16. Tannenholz, H., and Muir, K. B.: Arch. Path. **15**:789, 1933.

would many other substances injected intravenously, owing merely to their phagocytosing properties, or whether they have an important part in chemotherapeutic processes, cannot be said on the basis of the evidence at hand. The ability of arsine oxide to combine with sulphhydryl compounds suggests that the macrophages may be rich in these substances. The nitroprusside reaction was not positive, showing that these cells do not contain so high a concentration as does the epidermis, in which the reaction is positive. The finding of similar, although slightly different, granules in different portions of the lobules of the liver and at varying times after the injection of various drugs suggests that the liver handles each one in a different manner. The exact nature of the granules was not determined. They did not give the iron reaction, so they were not hemosiderin. They may have been a proteinate of the metal injected. The evidence at hand neither proves nor disproves the participation of the reticulo-endothelial cells in chemotherapeutic processes. Considerable study was made of the hydrogen sulphide method for demonstrating arsenic in tissues, and it was impossible to prove that the crystals demonstrated were arsenic tri-sulphide. They may have been a sulphur-protein compound.

#### SUMMARY AND CONCLUSIONS

Nondiseased white rats were given the following drugs intravenously or intramuscularly: neoarsphenamine, thio-arsene, colloidal bismuth, mercurosal, tryparsamide, gold sodium thiosulphate, sulpharsphenamine, bismuth arsphenamine sulphonate, potassium bismuth tartrate, mercuric chloride and sodium cacodylate. The dosages corresponded to about seven times the recognized therapeutic dose for adults. Half of the animals were given trypan blue, and half were left undyed. After the injection of neoarsphenamine, sulpharsphenamine, colloidal bismuth, mercuric chloride, gold sodium thiosulphate and thio-arsene, brownish granules were seen in macrophages in various parts of the lobules of the liver in sections stained with pyronine methyl green. After silver impregnation, granules were found irregularly after the injection of neoarsphenamine. No significant morphologic change was seen in the macrophages following the injection of the aforementioned drugs. No difference was observed between "blocked" and "unblocked" animals. This study shows that the reticulo-endothelial system undoubtedly participates in disposing of many drugs injected intravenously, but its actual function in chemotherapy is not definitely known. The reticulo-endothelial cells do not contain sufficient sulphhydryl compound to give a positive nitroprusside reaction, so that nothing definite can be said relative to their participation in the oxidation-reduction reaction attributed to those substances. Crystals found in tissues treated by the hydrogen sulphide method for demonstrating arsenic could not be proved to be arsenic.

# Laboratory Methods and Technical Notes

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## A NEW ELASTIC TISSUE STAIN

RAPID METHOD FOR ELASTIC TISSUE, CONNECTIVE TISSUE, FIBRIN AND AMYLOID, EMPLOYING CONGO RED

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As the significance of pathologic changes involving elastic tissue is increasingly evident, the use of a rapid and specific stain becomes essential. A technic is presented here which demonstrates elastic and connective tissue, fibrin and amyloid. The method depends on the use of congo red, which specifically stains elastic fibers bright red. In addition a slight modification of the procedure is offered for the preparation of permanent sections showing amyloid in a characteristic color.

### TECHNIC OF METHOD FOR ELASTIC TISSUE, CONNECTIVE TISSUE AND FIBRIN

1. Fix tissues in 10 per cent formaldehyde twenty-four hours or longer.
2. Cut frozen sections about 10 microns thick.
3. Wash thoroughly in tap water.
4. Place sections in the following stain for ten minutes: 4 per cent aqueous congo red solution (in 5 per cent sodium citrate), 8 cc., and glycerin (chemically pure), 2 cc.
5. Wash quickly in tap water.
6. Transfer sections to 1 per cent aqueous potassium iodide solution for ten seconds (rotating section in the solution).
7. Wash thoroughly in tap water.
8. Place sections in the following stain for from five to ten minutes:

Resorcinol .....	3.0 Gm.
Aniline blue.....	1.5 Gm.
Orange G.....	2.5 Gm.
Phosphomolybdic acid.....	1.0 Gm.
Distilled water.....	100 Cc.
9. Wash thoroughly in tap water in a large basin. Float sections onto slides and blot with filter paper very thoroughly.
10. Dehydrate in absolute alcohol for two minutes, changing alcohol twice (dropping bottle method).
11. Clear in pure xylene.
12. Mount in gum damar.

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This method gives a colorful, well differentiated preparation that keeps well when protected from direct exposure to light.

Elastic fibers appear bright red; fibrin and connective tissue, dark blue. Red blood cells are colored orange yellow.

Sections embedded in paraffin may be used satisfactorily, but require longer staining periods. The staining technic is the same as for frozen



Elastic tissue stained with congo red. A section of aorta shows syphilitic mesa-ortitis with disintegration of elastic plates. The red-stained elastic fibers in the section are seen as deep black in the drawing. *A* indicates the fibrous connective tissue of the adventitia; *B*, media, showing extensive destruction of the elastic fibers by areas of necrosis and cellular infiltration, and *C*, the intima, thickened by atherosclerotic changes.

sections except that it requires thirty minutes staining in the congo red solution and thirty minutes in the aniline blue solution.

If nuclear staining is desired in addition to differentiation of elastic tissue, use the technic for staining amyloid given herewith.

## MODIFIED TECHNIC FOR STAINING AMYLOID

1. Stain loose frozen sections in Harris' hematoxylin about fifteen seconds and wash thoroughly in tap water until sections are light blue.
2. Transfer sections to 4 per cent aqueous congo red solution with glycerin for fifteen minutes and wash in tap water.
3. Differentiate in 1 per cent aqueous potassium idoide solution and wash thoroughly in tap water.
4. Float sections onto slides and blot with filter paper.
5. Dehydrate in absolute alcohol for two minutes, changing alcohol at least twice.
6. Clear in xylene for from two to five minutes.
7. Mount in gum damar.

Amyloid is stained red and is beautifully differentiated from the surrounding tissues. The clearing of the tissue does not in any way interfere with the amyloid stain and the picture is further enhanced by the hematoxylin nuclear stain. The elastic fibers, also, are stained red, but are of a darker hue and may be easily differentiated from the amyloid.

# General Review

## EXPERIMENTAL CIRRHOSIS IN RELATION TO HUMAN CIRRHOSIS

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### CRITERIA

Medical literature abounds in reports on experimental cirrhosis. Probably no other pathologic condition has been produced by so many diverse methods. Before attempting to evaluate the bearing which these have on cirrhosis in man, it is necessary to consider the essential characteristics of human cirrhosis.

The conception that "cirrhosis" is synonymous with "chronic diffuse hepatitis" is generally accepted. Any agent or combination of agents which produces chronic diffuse hepatic inflammation will result in some degree of cirrhosis. The degree and character of this may vary but the essential features of the cirrhotic process are (1) degeneration and destruction of liver cells, (2) regeneration of liver cells from those which escaped destruction and (3) proliferation of connective tissue.

Portal cirrhosis<sup>1</sup> in advanced stages is accompanied by marked disturbances of the portal circulation. Kretz showed that normally the hepatic and portal veins divide in an arborescent fashion, and the smaller twigs are given off almost at right angles. The two venous systems may be compared to two huge venous trees the branches of which interlace regularly from opposite directions. They end in fine dendritic

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Prepared by assignment for the Second International Conference for Geographic Pathology, Utrecht, July 26-29, 1934.

1. Nomenclature: The term "*portal cirrhosis*" is recommended as synonymous with "*Laënnec's cirrhosis*." This avoids ambiguities implied in other terms and suggests the obstruction to portal circulation which is a regular feature in advanced stages of the disease. The term "*atrophic cirrhosis*" is objectionable because: 1. It implies atrophy, which is not regularly present; frequently the liver is larger than normal. 2. The term is used by some to indicate especially that form which develops following acute yellow atrophy. The term "*alcoholic cirrhosis*" suggests an etiologic agent whose rôle has not been demonstrated. The terms "*monolobular*," "*perilobular*" and "*multilobular*" are ambiguous since they are not used in the same sense by all writers. The terms "*nodular*," "*granular*" and "*hobnail*" cirrhosis describe a gross characteristic which may be present in conditions other than *portal cirrhosis*. The term "*Laënnec's cirrhosis*" is not objectionable. It has historic significance and is seldom misunderstood.

branches which dovetail alternately. The relationship of the two venous systems is clarified by considering the hepatic lobules in longitudinal sections instead of the cross-sections in which they are usually represented. This histologic aspect is shown diagrammatically in figure 1A. Such a

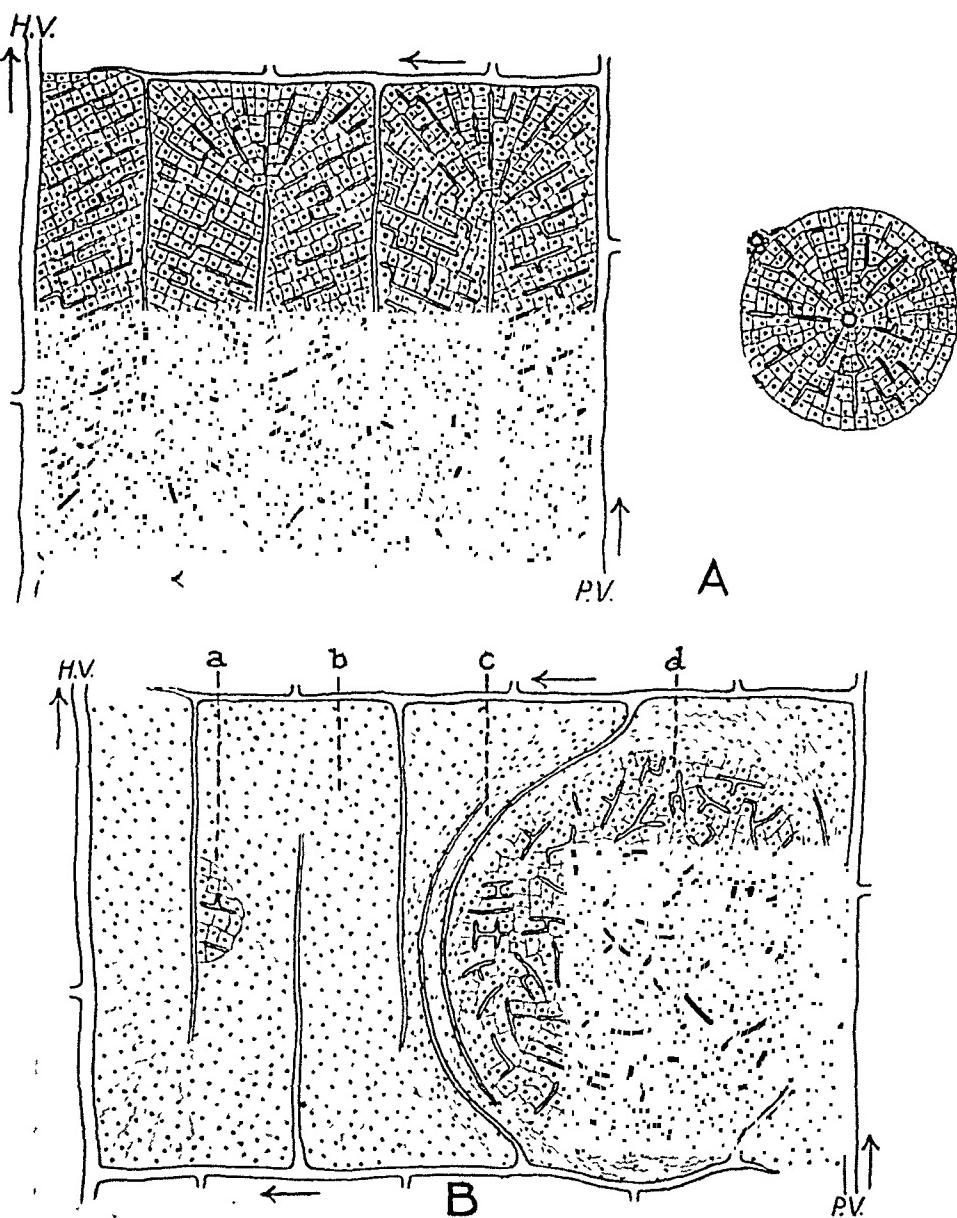


Fig. 1.—*A*, diagram of liver lobules in longitudinal section. This illustrates the alternating arrangement of the hepatic and portal venules more clearly than the conventional cross-section of a lobule. *P.V.* indicates the portal vein; *H.V.*, the hepatic vein.

*B*, diagram of liver lobules in longitudinal section, illustrating the development of cirrhosis: *b* represents an area of hepatic necrosis, and *a*, a group of intact hepatic cells, which by regeneration would produce an expanding nodule of cells, as *d*, and would compress the collapsed stroma and vessels about its periphery, *c*.

view of the circulation of the normal liver aids in visualizing the histogenesis of portal cirrhosis and its associated circulatory disturbances.

Alteration or destruction of architectural pattern has been noted as a prominent feature of portal cirrhosis by many who have studied this condition closely. Kertz believed that Laënnec's cirrhosis is the result of attacks of degeneration and destruction of liver cells, that the entire parenchyma has been totally reviewed several times, and that this results in a complete architectural transformation of the parenchyma. He found that the circulatory relations were markedly disturbed; that an injection mass introduced through the portal vein found its way directly into the hepatic vein leaving much of the parenchyma unpenetrated, and that there were many turgescent nodules of hepatic cells entirely devoid of hepatic veins. These nodules could be reached by injection through the hepatic artery but were largely impervious to injection via the portal vein.

Kelly believed that some injury caused destruction involving areas of liver tissue. As the substance of necrotic cells was absorbed the stroma of those areas collapsed. The expanding force of adjacent regenerating nodules of cells compressed this stroma and the vascular channels contained in it. He emphasized that the chief characteristic of Laënnec's cirrhosis is complete disarrangement of hepatic architecture. The regenerated nodules did not contain radicles of the hepatic vein. Both central and portal veins were diminished in number and were located at the periphery of the nodules. These interpretations and conclusions were later confirmed by McIndoe.

MacCallum gave a concise description of the cirrhotic process:

The liver-cells are killed in patches—whole lobules and groups of lobules at a time, or only parts of lobules. There remain irregular masses of liver tissue partly disconnected from their bile ducts. . . . The masses of liver-cells quickly increase in size by multiplication of their cells, new capillaries are formed in every direction, and this labyrinth of cells expands, pressing the stroma away on all sides. For a time the liver-cells are normal, but then comes another injury, and many of the hyperplastic nodules are partly destroyed. The whole process is repeated, and not only once, but many times. . . . It is clear that this must lead to an extraordinary distortion of the liver's structure. There are no longer lobules, but only nodules produced by the hyperplasia of smaller groups of cells which were left intact.

McNee expressed the same interpretation in very similar terms. Ghon's description of the essential histologic characteristics of cirrhosis presents the same view. He emphasized architectural alteration with complete disarrangement of the circulation as a prominent feature of Laënnec's cirrhosis. Mann maintained that alteration of architectural pattern is the distinguishing characteristic of Laënnec's cirrhosis. Moon's observations on active cirrhosis led to identical conclusions. In portal

cirrhosis the structural rearrangement is so marked that many observers have compared the nodules to adenomas. Laënnec himself regarded the nodules of regenerated hepatic cells as neoplastic. Figure 1B represents diagrammatically the development of this type of cirrhosis.

McIndoe's injection studies of cirrhotic livers showed a marked diminution in the total vascular bed. The main trunks were attenuated and irregularly stenosed. The larger branches were given off at unusually abrupt angles and showed irregular deviations to one side as if displaced by some invisible force. The expansive force of a growing mass of hepatic cells would produce such an effect. It was difficult to find any normal central veins whatever. The disarrangement of the terminals of the portal and hepatic veins was conspicuous. These terminals lay in distorted positions in the stroma surrounding the regenerated hepatic nodules. These nodules had no channels into which an injection could penetrate from the portal vein. He found that such nodules received blood principally through branches of the hepatic artery, the terminals of which are less easily obstructed than those of the portal vein. Thus in advanced cirrhosis a definite portohepatic venous obstruction develops, and the portal blood supply to the remaining parenchyma of the liver is reduced to a minimum, if it is not altogether absent. The principal cause for this circulatory block is the contraction of scar tissue about the terminal twigs of both the portal and the hepatic veins. This mechanism of obstruction explains the portal passive congestion and the ascites which are characteristic of late stages of cirrhosis.

Mann found that the capacity for regeneration is largely dependent on adequate portal circulation. When the portal blood is shunted into the vena cava by an Eck fistula, the regenerative capacity of the liver is markedly reduced. Absence of regeneration was noted following injuries to the liver in dogs with Eck's fistula. Similar injuries in normal dogs were followed by complete restoration. This indicates that in advanced cirrhosis obstruction to the portal circulation hinders regeneration and restoration of the hepatic parenchyma. Furthermore, the progressive increase of fibrous tissue which accompanies the cirrhotic process forms a rigid cage about the remaining parenchyma and by contraction this fibrous tissue limits mechanically the space which the hepatic cells may occupy. In areas adjacent to the capsule this limitation is less effective, and nodules of proliferating cells expand and bulge above the surface, producing the characteristic nodular surface. Interference with the portal circulation and the contracting cage of fibrous tissue hinder effective regeneration of hepatic cells.

Single injuries, or those which act on limited portions of lobules, produce a pathologic picture structurally different from that described.

This picture varies depending on the amount and location of the destruction and of the fibrous tissue proliferation. Occasionally a liver is found showing evidence of varying degrees of chronic diffuse hepatitis but without obliteration of lobules and without evidence of extensive progressive injury. Such cases give none of the clinical manifestations of portal cirrhosis, and the diagnosis of chronic hepatitis, or cirrhosis, usually results from postmortem evidence. Some inflammation and irregular fibrosis may develop wherever destruction of cells occurs. Occasionally the inflammation results in obstruction of bile ducts, producing the features of biliary cirrhosis. In other cases the inflammatory process involves portions of lobules irregularly and even surrounds single cells or small groups of cells. But the lobular pattern in such cases is still recognizable. These variations of chronic hepatitis do not fall readily into any proposed system of classification. The frequency of such cases is exemplified in a series of 10,016 autopsies reported by Mallory. In 590 cases of well marked cirrhosis he found 110, or 18.5 per cent, such atypical cases which he could not classify. Those who have studied chronic hepatitis extensively are less vigorous in their support of exact classification.

Numerous experiments have shown that the liver has a marvelous capacity for regeneration. When large portions of the liver are removed surgically no demonstrable deficiency of hepatic function results. Subsequent examination shows that the liver has regenerated to approximately its former volume. Such regeneration is accomplished by the formation of new lobules of normal size, structure and circulatory supply. The architectural pattern in such regenerated liver is not altered and is indistinguishable from that of the normal liver. When a single severe injury to liver parenchyma is produced by one of the known hepatic poisons, the injured areas often will be restored without fibrosis and without alteration of architectural pattern. The injury may have destroyed three fourths of the cells in each lobule, as shown by biopsy following the injury. Yet shortly they are replaced by multiplication of adjacent cells in the lobules. The architectural pattern is not affected, and fibrosis or other visible histologic changes may not have occurred. This type of result following a single chemical or toxic injury must not be expected to follow continued or repeated injury of similar nature.

In the process of repair following injury, all hepatic structures seem stimulated to proliferation. Not only do cells of the liver regenerate but there is also proliferation of bile ducts, of young fibrous tissue and of Kupffer cells and vascular structures. The numerous experiments in which cirrhosis has been produced experimentally have shown this fact conclusively. Hence proliferation of bile ducts is not a suitable differential criterion.

To recapitulate: The cirrhotic process consists of degeneration and destruction of hepatic cells, followed by regeneration and by fibrous tissue proliferation. It is essentially nothing else than chronic, progressive, diffuse hepatitis. If such destruction is limited in extent, involving only portions of the lobules, no marked structural or functional disturbance results. Biliary obstruction may occur, but there is no obstruction to the portal circulation or obliteration of hepatic architecture.

Varying degrees of minor injury without distortion of pattern lead to varying degrees of chronic hepatitis not easily classified. If accurate histologic classification of these were possible, it would serve no useful purpose.

If there is progressive or repeated destruction affecting extensive areas, the resulting regeneration of hepatic cells produces nodules of parenchyma with abnormal circulatory relations. The radicles of both the portal and the hepatic veins lie on the periphery of the nodule, and are embedded in stroma and proliferated fibrous tissue which, as it contracts, produces obstruction of the portal venules. In such a liver the normal lobular pattern is obliterated. The architecture is altered. There are no longer lobules but only nodules of cells divorced more or less completely from normal circulatory relations.

Many authors have emphasized *destruction of architectural pattern* as a feature of portal cirrhosis, but none have proposed to use it as a differential criterion. It will be so used in the following sections when some criterion is necessary to determine how closely experimental cirrhosis has approximated portal cirrhosis in man.

*Caution Required in Evaluating Experimental Cirrhosis in Rabbits.*  
—Many of the experiments on cirrhosis have been made on rabbits. These experiments are difficult to evaluate because rabbits frequently acquire chronic hepatitis with cirrhotic changes. Van Heukelom, von Kahlden, Klopstock, Fischler, Findlay and others have made such observations. Ophüls examined 50 rabbits collected from different sources; he found only 6 with livers that were not diseased. Coccidiosis with varying degrees of chronic hepatitis was found in the majority. Cirrhotic processes were found in some livers in which no coccidiosis could be demonstrated. In Ophüls' opinion, this throws grave doubt on many reports of an experimental production of cirrhosis. He regarded rabbits as entirely unsuitable for such experiments.

Smetana published a detailed study of the life cycle of *Eimeria*, the parasite causing coccidiosis, of its epidemiology and occurrence and of the various lesions and manifestations of coccidiosis. This disease is almost universal among rabbits, and produces varying degrees of chronic hepatitis. Sometimes an apparently normal liver is found following

mild infection, but often the healing process produces diffuse perilobular fibrosis. This may be accompanied by the formation of pseudolobules producing a picture indistinguishable from true cirrhosis. Smetana stated that it is practically impossible to exclude coccidiosis as the cause when a cirrhotic liver is found in a rabbit. Because of the prevalence of this disease, he urged that rabbits should not be used for experiments on cirrhosis.

These observations make necessary an attitude of extreme conservatism toward all reports of the production of cirrhosis in rabbits.

#### AGENTS USED IN THE PRODUCTION OF CIRRHOSIS

(Numerous agents have been used with more or less success in the production of cirrhosis. These may be grouped as inorganic chemicals, organic substances, serologic agents and infections. In many instances combinations of agents were found more effective than one of the agents alone.)

#### INORGANIC POISONS

(*Phosphorus*.—Numerous reports of the effects of phosphorus on the liver have been published. Its use began in the early days of experimental pathology and has continued to the present.)

Wegner gave small doses of phosphorus to rabbits by mouth and continued the treatment through many months. This produced marked interstitial proliferation with nodular granulations in the liver. The illustration shows the gross characteristics of Laënnec's cirrhosis. Microscopically, perilobular fibrosis is shown, but there is no evidence of a distortion of the architectural pattern.

Dinkler found increase in connective tissue, proliferation of bile ducts and visible evidence of regeneration of hepatic cells following chronic phosphorus poisoning. Krönig compared the effects of acute and prolonged phosphorus poisoning in rabbits. Untreated litter mates of treated rabbits served as controls. Animals that died early showed only fatty degeneration and necrosis of liver cells. Beginning interstitial fibrosis, proliferation of bile ducts and evidences of regeneration of hepatic cells were found following phosphorus poisoning of longer duration.

Ackermann produced chronic phosphorus poisoning in rabbits. This was followed by perilobular connective tissue proliferation seen both macroscopically and microscopically. He regarded cirrhosis as the result of a defensive reaction of the tissue tending to counteract the effects of an injury—an early statement of a modern conception.

Aufrecht found diffuse inflammation and fibrous proliferation following phosphorus poisoning. The drawings that illustrate the article do not show whether a distortion of lobular pattern resulted.

De Josselin de Jong found periportal fibrosis, cellular infiltration and slight increase in the interstitial connective tissue in one rabbit which had been treated one hundred and twenty-nine days with phosphorus. Stolnikow reported degenerative changes in the liver followed by regeneration. There was no evidence of chronic hepatitis. Tischner found interstitial proliferation following phosphorus poisoning of long duration.

Paltauf reported 8 cases of phosphorus poisoning in man. The changes described in the livers of those who died between seven and fourteen days after the onset resembled closely those seen following death from acute yellow atrophy. Microscopically there was extensive parenchymatous and fatty degeneration, most marked about the periphery of the lobules. He noted visible evidences of hepatic cell regeneration from the biliary canals. There was young fibrous tissue proliferation in the perilobular spaces. He interpreted these changes as characteristic of early acute cirrhosis.

1) Fischler instituted an elaborate series of experiments on dogs, using a combination of phosphorus with amyl and ethyl alcohol. He found phosphorus combined with alcohol much more effective than phosphorus alone. 1) He made a study of the function of the liver throughout these experiments by means of a complete biliary fistula. The presence of bilirubin or of urobilin was used as an indication of a disturbance of hepatic function. The treatment produced varying degrees of hepatic degeneration and secretional disturbance, resulting in marked fibrous tissue proliferation in the periphery of the lobules and leukocytic and mononuclear infiltration. He interpreted the fibrous proliferation and evidences of inflammation as a reparative process following injury. Photomicrographs show marked perilobular fibrosis and infiltration, with the lobular pattern distorted but not obliterated.

1) (Balan found marked fatty changes in the liver but no cirrhosis following the use of phosphorus.)

(Mallory produced marked cirrhosis in rabbits and guinea-pigs by giving daily minute doses of yellow phosphorus with their food.) After five months the livers were scarred, contracted and nodular, the condition closely resembling Laënnec's cirrhosis. Microscopically the cytoplasm showed the hyaline degenerative changes which according to Mallory are characteristic of "alcoholic" cirrhosis. He suggested that phosphorus as an impurity in iron or tin containers might, by the action of acids, become dissolved in alcoholic liquors. However, he failed to find any phosphorus in 25 samples of spirituous liquors tested. He stated that after thirty-six years of experimentation on alcoholic cirrhosis he, like other investigators, had ruled out ethyl alcohol as the cause. He had attempted unsuccessfully to produce cirrhosis with over 30 toxic substances which might possibly be present as contaminants in

alcoholic liquors. Photographs of the livers of the guinea-pigs present the marked nodular, granular, contracted features of Laënnec's cirrhosis (fig. 2). The photomicrographs show marked perilobular cirrhosis. There is moderate extension of connective tissue into the margins of the lobules but no distortion or obliteration of the lobular pattern.

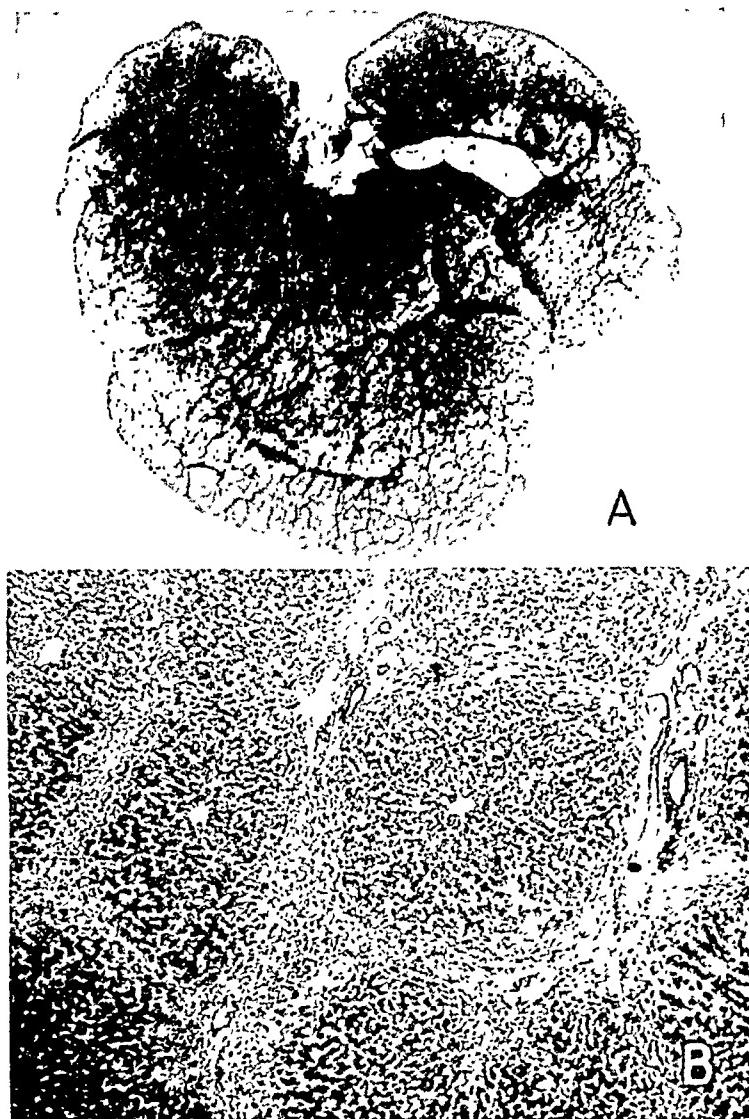


Fig. 2.—*A*, cirrhosis produced by phosphorus (Mallory). The gross features are indistinguishable from portal cirrhosis. *B*, photomicrograph of cirrhosis produced by phosphorus (Mallory). The cirrhotic process is marked, but the lobular pattern is not distorted. This does not reproduce the essential histologic feature of portal cirrhosis.

*Arsenic.*—One of the characteristic effects of arsenical poisons is the injury produced in hepatic cells.) Necrosis results if the injury is severe. Ziegler and Obolonski reported the effects of small doses of

arsenic on dogs and rabbits. In dogs only parenchymatous changes were seen. Podwyssotski found localized areas of necrosis in acute arsenical poisoning. Following chronic poisoning he saw proliferation of fibrous tissue. Wolko gave a solution of potassium arsenate subcutaneously to rabbits in experiments extending to forty days. He found marked proliferation of hepatic cells and bile ducts accompanied by leukocytic infiltration and proliferation of connective tissue in the periphery of the lobules and within the lobules.

Ehrlich made toxicologic studies on a huge series of arsenical compounds. He observed that degeneration and necrosis of hepatic cells were constant features of arsenical poisoning. This has been confirmed by Grote, Fischler, Sollmann and others. Fischler believed that a combination of arsenic with alcohol would prove as effective as phosphorus with alcohol in producing cirrhosis.

(Only a few experiments on the pathologic effects of organic arsenicals have been found.) Testoni gave from 1.5 to 10.58 cc. of a 10 per cent solution of solarson to rabbits by intravenous injection. The animals died in from two to one hundred and fourteen hours. In some of the livers there were striking parenchymatous changes associated with diffuse hemorrhage. In others only congestion and hemorrhage were seen. Heitzmann gave acetarsone (stovarsol) and treparsol (a product similar to acetarsone) in amounts of from 0.5 to 1.5 Gm. by mouth to rabbits and guinea-pigs. The animals were killed from three to sixteen days later. Diffuse fatty changes of liver cells and Kupffer cells resulted. Chronic effects were not studied.

It has been observed repeatedly (Kuczynski, Gilberg and Föckler, Choisser and Wilson, Meyer and Heubner, Bortin, McDonald and others) that arsphenamine and similar arsenicals occasionally produce acute poisoning in susceptible persons. When death results in such cases, severe necrosis of the liver is found. Often the condition of the liver is described as resembling acute yellow atrophy. (O'Leary, Snell and Bannick reported two cases of portal cirrhosis following chronic arsenical poisoning.) They regarded arsenical cirrhosis as a definite clinical entity of which their cases were representative. They cited similar phenomena recorded by other observers. (Rolleston and McNee stated that arsenic appears to be capable of setting up cirrhosis of the liver.

*Lead.*—Litton reported necrosis of the liver following injection of lead chromate into the mesenteric vein.

Coen and D'Ajutolo reported early stages of interstitial hepatitis in rabbits following the use of lead acetate. This was accompanied by a hyperplastic pericholangitis with fibrous tissue proliferation. Potain, Prevost and Binet, and Annino found localized necroses and some inter-

stitial proliferation following chronic lead poisoning. Lafitte reported 6 cases of cirrhosis in man following chronic lead poisoning, with no history of alcoholism. He produced chronic lead poisoning in 4 rabbits. The experiments lasted from forty-two days to five months. Degeneration and necrosis of hepatic cells were followed by perilobular connective tissue proliferation and atrophy of hepatic cells. The livers were indurated and contracted. Albot gave lead carbonate and lead acetate subcutaneously and by mouth to rats, guinea-pigs and rabbits. Degenerative changes of hepatic cells followed by reticulosclerosis resulted. In lead intoxication of long duration, he described a form of diffuse hepatitis with periportal fibrosis and atrophy of cells in central areas.

Lancereaux believed that potassium salts used in the manufacture of alcoholic beverages might be responsible for cirrhosis. He fed potassium sulphate and potassium bisulphate to dogs, rabbits, guinea-pigs and rats. The conditions and results of the experiments are not clearly stated. He argued from experimental evidence and clinical observations that wines which had been cleared with calcium sulphate were more productive of cirrhosis than other liquors. This explanation was refuted by Vallin, Rendu and others, whose observations did not corroborate those of Lancereaux. Clinical observations on cirrhosis related to alcoholism are not limited to the use of "plastered" wines. Spirituous liquors have been incriminated more than wines. Lancereaux' observations have lacked experimental confirmation.

Gye and Purdy gave intravenous injections of sublethal doses of colloidal silica to rabbits. Poisonous doses produced parenchymatous degeneration and necrosis in the liver, spleen and kidneys. Chronic poisoning with smaller doses produced extensive cirrhosis. The descriptions are confirmed by photographs, but neither the histologic descriptions nor the illustrations indicate that distortion or obliteration of lobular pattern resulted from the effects of colloidal silica.

Polson gave iron for periods of from three to four years to adult rabbits, maintaining a very high level of iron in their organs. No cirrhosis or pancreatic damage was produced. He concluded that it is unlikely that excess of iron is responsible for the hepatic and pancreatic changes found in hemachromatosis.

Huguenin, Nemours-Auguste and Albot studied the effects of thorium dioxide in a series of rabbits. A single large dose caused death in a few hours, with marked necrosis of hepatic cells. Smaller doses administered over a period of time resulted in various stages of hepatitis. In the more chronic cases this produced a form of cirrhosis. Deficiency of hepatic function was demonstrated by the galactose test. The illustrations show periportal fibrosis without disturbance of architectural pattern.

Scott and Helz fed sodium aluminum sulphate to 80 white rats in concentrations up to 2 per cent of their food. Protracted feeding produced no deleterious effects on growth or reproduction. The livers appeared normal and contained a normal amount of iron. No abnormal gross or microscopic features were seen in the liver or in other organs.

Bolliger and Inglis found that a single treatment with roentgen rays produced pathologic changes in the livers of dogs surgically exposed. The immediate result of such treatment was necrosis of hepatic cells, not only superficially but also in the deeper portions though in less degree. In the animals which survived such treatment jaundice and ascites developed, and examination of the livers showed proliferation of connective tissue.

*Manganese.*—Findlay gave repeated small doses of manganese chloride by injection or by mouth to rabbits, guinea-pigs and rats. Acute poisoning resulted in degeneration and necrosis of cells about the periphery of the lobules. Subsequently cirrhotic changes occurred which were first recognizable as fibrosis about the periphery of the lobules. Later the proliferating fibrous tissue invaded the lobules. The cirrhosis was monolobular, and Findlay regarded it as biliary in type. Jaundice occurred frequently. His numerous illustrations confirm the descriptions. Marked invasion of the lobules by fibrous tissue is shown. In some instances the lobules are distorted, but the lobular pattern is still recognizable.

Handovsky, Shultz and Staemmler treated rabbits with manganese chloride and with organic salts of manganese. These regularly produced degenerative changes of the hepatic cells and round cell infiltration. In some there was fibrous thickening about Glisson's capsule, which they interpreted as the effects of chronic poisoning, although they made no claims to the experimental production of cirrhosis.

Martin made similar experiments with manganese chloride on guinea-pigs. His attention was directed particularly to early minute cellular changes indicated by alterations in the chondriosomes. He emphasized the importance of these as indicators of early cellular damage. His experiments led to the conclusion that parenchymatous lesions precede interstitial changes in the cirrhotic process.

Hurst and Hurst studied the effects of manganese chloride, chloroform and phenylhydrazine hydrochloride on the livers of guinea-pigs and rabbits. Subcutaneous injections of any one of these substances in quantities of from 0.15 to 0.25 Gm. per kilogram caused severe intoxication ending in death within twenty-four hours. In such cases extensive necrosis of the liver was present. Smaller doses repeated three or four times weekly produced extensive fibrosis of the liver. With chronic manganese poisoning cirrhosis developed more readily in rabbits than

in guinea-pigs. The condition of the liver was similar to human monolobular cirrhosis. A chronic inflammatory reaction was evidenced by cellular infiltration and by proliferation of bile ducts and connective tissue. Photomicrographs of such livers show that the lobular pattern was not destroyed, but in some instances the fibrosis was rather diffuse throughout the lobules. The injection of living colon bacilli into the portal circulation during the course of manganese poisoning in guinea-pigs markedly increased the severity of the cirrhotic process. Fibrosis was stimulated, resulting after a comparatively short time, in an intense cirrhosis resembling that of Laënnec. They found also that injections of manganese chloride were more effective in producing cirrhosis when combined with injections of phenylhydrazine. Photomicrographs of the livers following this treatment show unmistakable distortion and destruction of lobular pattern.

One object of their experiments was to observe degenerative changes in the brain, especially those in the lenticular nucleus, associated with hepatic cirrhosis. No such changes were found in their animals.

Rao found that manganese fed to rabbits led to necrosis in the periphery of the lobules in cases of acute poisoning. Small doses repeated over a period of time resulted in cellular proliferation, increase in collagen fibers, and other inflammatory changes in the periportal connective tissue. This produced a form of monolobular cirrhosis which he did not regard as similar to Laënnec's cirrhosis.

Albot found degeneration and proliferative changes having the nature of a diffuse hepatitis in guinea-pigs and rabbits following injections of manganese chloride subcutaneously. Van der Schueren reported marked degeneration of hepatic cells and slight periportal sclerosis in manganese chloride poisoning in guinea-pigs.

*Copper.*—Mallory, Parker and Nye reported the effects of prolonged feeding of copper acetate to animals. They found that this was followed by a chronic lesion of the liver in which heavy pigmentation was combined with cirrhosis. Rabbits and rats were more susceptible than guinea-pigs and monkeys. In from six to twelve months the condition of the liver in the rabbits resembled human hemachromatosis. Three cases of cirrhosis developed among 22 rabbits treated with copper. They believed that copper contained in food or drink is an important cause of hemachromatosis, and that alcohol accentuates the effects of copper.

Hall and Butt gave copper acetate by mouth or by injection to rabbits, rats and sheep, and found that it caused a marked increase in the copper content of the liver. They reported that the storage of copper in the liver was paralleled by a storage of hemofuscin which contained iron in a masked form. This, they and Mallory believed, was slowly

converted into hemosiderin, perhaps after some years. Alcohol given with copper acetate did not hasten or increase the deposit of pigment. Pigmentation was followed by fibrosis, producing a condition resembling early hemachromatosis. This was more marked in rabbits than in other animals. They did not believe that copper occurs in human food or drink in sufficient quantities to produce this effect.

Flinn and Von Glahn attempted the production of pigmentary cirrhosis by giving copper, but without success. They attributed the pigmentation reported by others in such experiments to the deposit of pigmented lipoids derived from exogenous sources such as vegetables in the diet, especially carrots. They found no difference in animals given sodium acetate as compared with those given copper acetate.

Polson gave copper acetate to rabbits as described by Mallory. He found 1 rabbit with cirrhosis among 18 that were treated with copper. A control group were fed on mangel-wurzels and turnips and given no copper. He reported a deposition of hemofuscin in the liver in 87.5 per cent of these.

Adrianoff fed rats organic and inorganic copper compounds. One series of rats were given from 1 to 20 mg. of copper sulphate in their daily diet. After from nine to fifteen months several of the animals showed an annular type of cirrhosis with proliferation of bile ducts. Animals fed copper plus alcohol or copper plus olive oil showed no greater tendency to cirrhosis than those fed copper alone. Other groups of animals were fed copper acetate, copper oleate, copper-lead stearate copper-aluminum oleate, aluminum stearate, iron stearate and bismuth stearate. One rat fed lead stearate showed moderate cirrhosis. No cirrhosis was found in any of the others in the series. He believed the inorganic salts of copper were more effective than the organic compounds.

Oshima and Siebert fed copper sulphate to rabbits over a period of eight months. The livers of these animals contained about sixteen times the normal content of copper. No cirrhosis was produced.

Herkel reported the effects of prolonged feeding of various copper salts to rabbits and rats. This produced storage of copper in the liver but no demonstrable evidence of pigmentation or of cirrhosis.

Mallory and Parker extended their former series of experiments and demonstrated that copper is stored in the liver and may be demonstrated there within twenty-four hours after its injection. They used guinea-pigs, rabbits, monkeys and sheep, and injected a 20 per cent suspension of metallic copper in lard subcutaneously. They found that acute poisoning with copper produced anemia, hemoglobinuria, pigmentation and necrosis of hepatic and renal cells. They believed that the

pigmentation of the liver was derived from the destruction of hemoglobin. Repeated injections produced a form of pigmentary cirrhosis resembling hemachromatosis.

Schindel fed copper sulphate with and without alcohol to rabbits and rats. There was marked accumulation of copper in the livers in experiments lasting one hundred and seven days, and in longer experiments there was pigmentation of the Kupffer cells. No cirrhosis resulted.

Hall and MacKay confirmed their previous results and those of Mallory, Parker and Nye by a carefully controlled study of the effects of copper acetate in rabbits. For each animal that received copper, a litter mate of the same sex was used as a control. It was given the same diet with copper omitted; vegetables containing carotene were excluded from the diets. Twenty-one rabbits received a diet containing 2 mg. of copper acetate per gram of food. The experiments lasted from twenty-one to one hundred and five days. Seventeen of the copper-fed animals showed hemofuscin pigmentation in the Kupffer cells, and 9 showed, in addition, cirrhosis of the liver. Five showed varying degrees of necrosis but no cirrhosis. None of the control animals for this group, and none of another group of animals receiving sodium acetate instead of copper acetate, showed pigmentation or cirrhosis. The cirrhosis was perilobular, and the lobular architecture was not obliterated.

In another experiment 22 rabbits were fed diets rich in carrots, from thirty-two to fifty days. Three presented Kupffer giant cells which contained a moderate amount of hemofuscin, 1 early cirrhosis, and 2 slight connective tissue proliferation. This, perhaps, is not a higher occurrence of hepatic changes than should be expected in rabbits.

Villaret, Bertraud, Justin-Besançon and Even injected acetates of cobalt, copper, manganese and nickel into guinea-pigs and rabbits. The livers showed degenerative changes followed by a gradual increase in fibrous tissue. The sclerosis was evident about the third or fourth month. Pigmentation was not noted in this series.

It appears that many inorganic poisons are capable of producing degeneration and necrosis of hepatic cells, and that the prolonged effects result in reparative processes of a cirrhotic nature. Moderate distortion of lobular pattern was produced in animals other than rabbits by the use of phosphorus plus alcohol (Fischler) and by manganese chloride (Findlay). Marked distortion of pattern was produced by Hurst and Hurst with a combination of injections of manganese chloride and intraportal injection of colon bacilli. The relationship between copper and pigmentary cirrhosis is not clearly established, since there is absence of general agreement in the evidence. It seems improbable that inorganic poisons, with the possible exception of arsenic compounds, constitute an important factor in the etiology of human cirrhosis.

## ORGANIC SUBSTANCES

A wide variety of organic chemicals and substances of protein composition have been used experimentally with resulting damage to the liver. Immune serums and anaphylactic reactions have resulted in necrosis of hepatic cells. In some instances such damage has been slight; in others it has resulted in chronic inflammatory changes. Varying degrees of fibrosis up to marked cirrhosis with obliteration of lobular pattern have been reported.

*Alcohol.*—The belief that cirrhosis is due to alcoholism has led to numerous experiments on the effects of alcohol. For a complete summary of experiments made prior to 1911, the reader is referred to reviews by van Heukelom, Klopstock, Poggenpohl, Saltykow and Fischler. Rössle's recent treatise on inflammations of the liver presents a conservative summary of experimental results from the use of various agents including alcohol. The following results are summarized from the review of Saltykow, whose attitude is sympathetic toward the experimental production of cirrhosis with alcohol. Dahlstrom Duchek, Joffroy and Serveaux von Baumgarten and Reiter<sup>2</sup> found no effects in animals following treatment with alcohol.

Kremiansky, Ruge, Challand, Pupier, Strassmann, Afanassijew, Kublin, Magnam, Mairet and Combemale, Tobaldo, von Kahlden, Ing hilleri, Jovine, Friedenwald, D'Amata, Lafitte, Dujardin-Beaumetz and Audige, and Grandmaison<sup>2</sup> found varying degrees of degenerative changes with or without slight or moderate infiltration about the portal structures. Most of these inflammatory changes were found in rabbits. Those credited with producing marked interstitial proliferation, amounting to cirrhosis, were Strauss and Blocq, De Rechter, Mertens, Saltykow and Fahr. The results obtained by these workers, whose reports are quoted frequently, and the experiments subsequent to 1911, will be reviewed briefly.

Strauss and Blocq gave mixtures of ethyl and amyl alcohol daily to 24 rabbits through a stomach tube. Most of the rabbits died within three months. Parasitic lesions were noted in the livers of several. There were no other significant findings. Two lived seven and one-half and twelve months, respectively. Both showed marked perilobular lymphocytic infiltration and proliferation of young connective tissue, which was interpreted as early cirrhosis. The illustrations indicate chronic periportal inflammation; the lobules appear normal, and there is no distortion of pattern.

De Rechter used 10 rabbits and 3 dogs. These were given a mixture of ethyl alcohol, 22.5 per cent, amyl alcohol, 2.5 per cent, water, 75 per

2. For references to the original articles of some of these authors, the reader is referred to the reviews by Fischler, Saltykow, and others.

cent, by stomach tube. Several animals died early in the experiment. The report is based on 2 rabbits which lived five and a half and nine months, respectively, and 1 dog which lived twenty weeks. The livers of the rabbits were described as firm, fibrotic and granular with a peri-lobular increase in connective tissue. Periportal fibrosis was seen in the dog's liver. The report is indefinite and is illustrated with diagrammatic drawings. These do not show distortion of lobular pattern, and the evidence of cirrhosis is inconclusive.

The report of Mertens is frequently misquoted, and in many libraries his original article is not available, hence special attention is given to his experiments. Mertens gave chloroform mixed with paraffin by subcutaneous injection to 15 rabbits. The doses ranged from 0.25 to 0.5 cc. of chloroform given at intervals of from three to ten days. The experiments extended from six days to three and one-half months. Parasitic lesions of the liver were noted in several rabbits. The acute effects of chloroform on hepatic cells were marked granular degeneration, vacuolation, fatty degeneration and chromatolysis. The chronic effects were cirrhotic. He concluded that the results proved indisputably "that the administration of chloroform in small doses continued over a long time produced in rabbits a disease presenting all the characteristics of Laënnec's granular atrophy of the liver in man."

In another experiment 12 rabbits lived from twenty-five days to eleven months in an atmosphere charged with alcohol by evaporation. Several died early with evidences of parasitic disease. Details are given of the gross examination of each rabbit, but the microscopic observations are recorded in general, without reference to individual animals. He stated that in some the liver was firmer than normal, that there was an abnormal proliferation of connective tissue in the portal spaces, and that ascites was abundant. One notes that in 5 of the 12 rabbits there was fluid in the peritoneal cavity described as clear in some and as flocculent or purulent in others. In several instances the presence of this fluid coincided with the presence of parasitic lesions in the liver. In no instance was the fluid associated with a liver having cirrhotic characteristics. He found varying degrees of cellular degeneration and some inflammatory reaction accompanied by leukocytes and proliferating fibrous tissue in the periportal areas. He made no claim for the production of cirrhosis by alcohol.

The article is illustrated by thirteen figures, eleven of which show the effects of chloroform and support adequately the author's conclusion that they resemble Laënnec's cirrhosis. For the animals which received alcohol only two drawings are presented. These show hepatic cells in high magnification and illustrate degenerative changes only. In one a multinucleated giant cell is represented. No proliferation or inflamma-

tory changes are shown. Parasitic disease was prevalent among Merten's rabbits. In only 2 of the alcohol-treated rabbits was it stated that no parasitic lesions were seen. Disregarding coccidiosis, one would conclude that the experiment with chloroform resulted in marked cirrhosis. Likewise one would conclude that in the experiment with alcohol no acceptable evidence of cirrhotic change was shown.

Saltykow gave minute doses of alcohol diluted with saline solution intravenously to 3 young rabbits. Two died soon and their organs showed nothing significant. The third rabbit lived two years and seven weeks, having received in eight doses a total of 2.5 cc. of absolute alcohol. One month following the last injection the rabbit became severely ill and died fourteen days later.—Marked cirrhosis and arteriosclerosis were found in this rabbit.) Apparently Saltykow regarded this experiment as significant.

Fahr fed alcohol to 3 rabbits and 2 guinea-pigs for periods extending to three years. Marked fatty changes of the liver resulted. The liver of 1 rabbit showed fibrosis, but Fahr was not convinced that it should be interpreted as cirrhosis.) He stated: "The cardinal symptom of chronic alcohol poisoning is fatty change of the liver. This may or may not be accompanied by inflammatory changes."

The details of one significant experiment with negative results will be given. Under the auspices of the Committee of Fifty, Friedenwald made an extended investigation on the effects of alcohol in animals. This was given in the form of whisky or of absolute alcohol suitably diluted, by stomach tube, to 120 rabbits. Acute alcoholic poisoning lasting a few days and chronic effects of more than four years' duration were studied. One rabbit received 14,925 cc. of absolute alcohol. Each of 49 animals received more than 1,000 cc. of alcohol. One rabbit received 18,751 cc. of whisky. Twenty-three rabbits received over 1,000 cc. of whisky. A great majority of the animals showed fatty changes of the liver, heart and kidneys. These effects were temporary. They were not found in the animals whose treatment with alcohol had been discontinued a short time prior to their examination. In 1 rabbit cirrhosis with ascites was found; this was not regarded as significant in so large a series of animals. The evidence from this experiment was reviewed and verified by William H. Welch, who agreed with Friedenwald in the conclusion that such lesions as cirrhosis, chronic nephritis or arteriosclerosis had not resulted.

The review by Saltykow would be misleading to those unfamiliar with the original reports. It contains inaccuracies of statement, and the reviewer speaks *ex parte*. An attitude of objectivity toward the evidence is lacking. Fahr is credited with producing cirrhosis, which Fahr did not claim. Merten's results are interpreted as the successful

production of cirrhosis with alcohol and are defended with argument. Saltykow's result with 1 rabbit is stressed, but no mention is made of the conditions and results of Friedenwald's experiment on 120 rabbits. Coccidiosis as a factor in various experiments in which cirrhosis occurred is lightly set aside. The review summarizes the effects of alcohol on various organs and emphasizes its cirrhotogenic action.

(Bischoff gave whisky in quantities of from 10 to 20 cc. daily to young rabbits through a stomach tube.) The treatment continued from three to seven weeks. Marked fatty infiltration of the heart and liver resulted. This was especially marked in the central portion of the lobules. The kidneys showed no effects. (No changes resembling cirrhosis were found.)

(In Schafir's experiment, 22 rabbits received whisky by stomach tube; 9 rabbits received weekly intravenous injections of from 0.1 to 1 cc. of absolute alcohol suitably diluted; and 10 rabbits, receiving whisky by stomach tube, were given egg yolk and milk in their diet. Degenerative changes and fatty infiltration occurred in the first group, also localized areas of connective tissue proliferation and infiltration by small cells. The statement that these changes occurred in individual animals without relation to the length of treatment indicates that some process not related to the treatment caused them. The second group showed similar changes, described as more marked, and in the third group fatty infiltration predominated. Biopsy specimens of the liver were taken from a few rabbits in each group prior to treatment, for comparison of the condition before with that subsequent to treatment. Most of the rabbits died of intercurrent infections. It was not stated how many died, how long treatment of the survivors was continued, or how much alcohol was given. The observations were recorded in general, without reference to individual animals. Fatty changes and increase in stroma similar to those found in the liver in alcoholism were described. (The author stated that no marked interstitial changes resulted. Photomicrographs at high magnification illustrate degenerative and fatty changes, and do not show cirrhotic characteristics.)

(Lissauer reported the results of injection of 50 per cent alcohol and of brandy intravenously into rabbits.) The number of animals and the duration of treatment were not stated. In 1 rabbit the liver was firm and nodular. (Microscopically there were infiltration and fibrosis about the markings of the lobules.) No illustrations were given. He refrained from drawing conclusions from this experiment.

(Kryle and Schopper gave alcohol intravenously, subcutaneously and by mouth to 34 rabbits.) The rabbits lived from one day to thirteen weeks. Parenchymatous degeneration occurred generally. Marked fatty degeneration was seen in 10. Slight infiltration by small cells was noted in 7;

in 6 this was outstanding and was accompanied by proliferation of connective tissue interpreted as "beginning cirrhosis." In 3 they regarded chronic inflammation as "genuine cirrhosis." Many animals died early. The rabbit which survived longest received in thirteen weeks 870 cc. of 50 per cent alcohol. (The liver showed beginning cirrhosis.) Drawings furnished as illustrations show degenerative changes and varying degrees of chronic perilobular inflammation.

(Isobe treated 80 rabbits and 7 dogs with alcohol and potassium sulphate, singly and in combination. The experiments extended from one to twenty months. In many of the animals there were severe parenchymatous degeneration and fatty infiltration.) A few showed

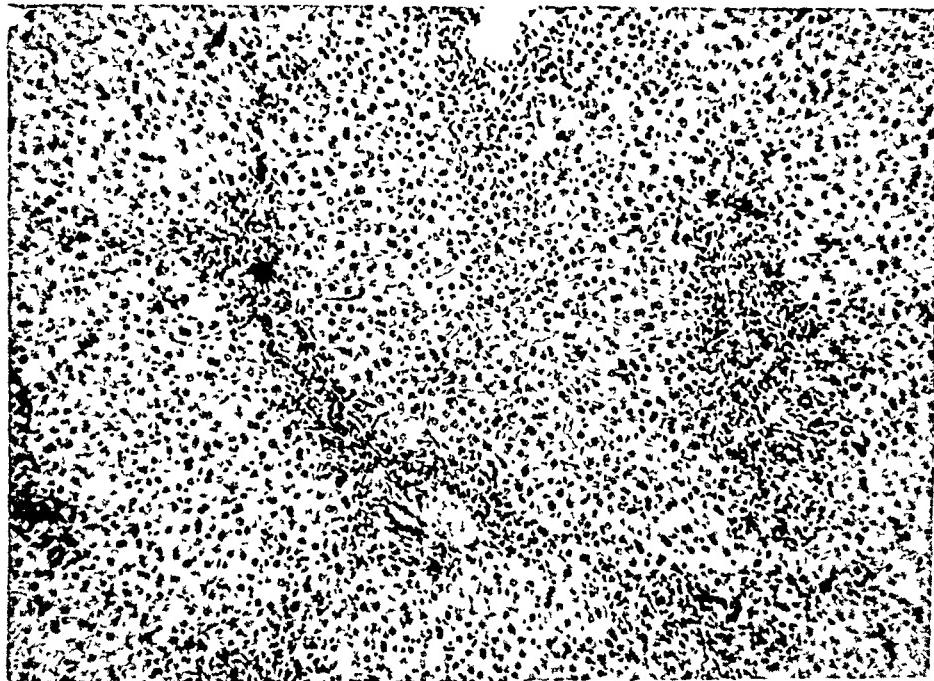


Fig. 3.—Photomicrograph of liver from an untreated rabbit. This type of chronic periportal inflammation is common among adult rabbits. It may easily be mistaken for "beginning cirrhosis."

round cell infiltration and moderate fibrosis of Glisson's capsule. The descriptions indicate a moderate chronic periportal inflammation. No illustrations are given.

(Grover fed alcohol to rabbits for periods of from three to twelve months. He reported cirrhosis in varying degrees in 6 of the 12.) His description of the microscopic changes indicates varying degrees of infiltration and of fibrous proliferation about the portal canals. The photomicrographs show moderate perilobular fibrosis such as is often seen in old rabbits (fig. 3). There is no distortion of lobular pattern. He credited Fahr with producing cirrhosis with alcohol, misquoted the

results of Afanassijew, Mertens and others, disregarded the experiment of Friedenwald, and concluded that cirrhosis can be obtained with alcohol independent of other factors.

Ogata gave alcohol to rabbits by injection into the portal vein and into the ear veins. In some of the animals there was perilobular fibrosis accompanied by degeneration of hepatic cells. No conclusions were drawn.

Wallace made clinical studies on cases of marked alcoholic intoxication. In 17 such cases the van den Bergh test for hepatic function showed a marked increase in urobilinogen and bilirubin. Urobilinogen was found also in the urine. He concluded that in acute alcoholic poisoning there is evidence of hepatic damage proportionate to the severity of the poisoning.

Rosenthal's experiments did not confirm Wallace's results. He found no increase of urobilin in the urine or of bilirubin in the blood following acute alcoholic poisoning in dogs. He found, however, that alcohol greatly increases the severity of chloroform poisoning in dogs. A two hour period of anesthesia from chloroform was fatal to 6 of 10 dogs previously intoxicated with alcohol given by mouth in a dosage of 2 cc. per kilogram of body weight. The livers of these dogs showed marked diffuse necrosis resembling that of acute yellow atrophy in man. In 10 dogs anesthetized two hours with chloroform but given no alcohol no fatalities resulted.

Adrianoff and Ansbacher mixed alcohol with food given to rats. The alcohol was in a concentration of from 20 to 50 per cent. In experiments lasting from nine to fifteen months no evidence of cirrhosis resulted. Van der Schueren gave 50 per cent alcohol in doses of 2 cc. per day to guinea-pigs during a period of from fourteen to seventeen months. He commented on the scarcity of resulting lesions. Two of the animals showed fatty degeneration of the liver. In 1 guinea-pig slight increase of connective tissue and mononuclear infiltration about the portal structures were seen.

MacNider reported hepatic changes resulting from acute alcoholic intoxication in dogs. Degenerative changes were produced in the cells in the outer zone of the lobules. These consisted of edema accompanied by an accumulation of lipoids. No necrosis resulted. In prolonged intoxication all portions of the lobules were involved. The cells returned to normal following such intoxication. Chronic effects were not studied.

(No cirrhosis has been reported in experiments with alcohol in animals other than rabbits. Several authors stated that in dogs, rats guinea-pigs, etc., the liver is much more resistant, basing their conclusion on the results in rabbits. Some even claimed that rabbits are especially

suitable because of the ease with which fibrosis can be produced in their livers!) Many of the illustrations and descriptions indicate the type of chronic periportal inflammation often found in the livers of old rabbits (fig. 3). Frequently the acknowledged presence of parasitic lesions in the livers was disregarded. Some authors have shown a non-critical attitude toward their results, and a few have appeared in the rôle of prosecuting attorney rather than in that of unprejudiced judge.

(Without minimizing the contributory or predisposing influence which alcohol may exert, it must be concluded that experimental evidence has not substantiated the belief that alcohol is a direct cause of cirrhosis. Combination of alcohol with other agents has greatly increased their toxic effects on the liver.) Fischler's experiments with alcohol and phosphorus, Scagliosi's results with alcohol plus bacteria, Wallace's results with alcohol plus chloroform and the results of Lamson, Wing, Mann and others with alcohol plus carbon tetrachloride are instances of the contributory effects of alcohol. It is probable that such influence as alcohol exerts in causing human cirrhosis results from its action in reenforcing or accentuating the effects of other agents or in producing degenerative changes in the hepatic cells, thereby rendering them more susceptible to injury.)

*Chloroform.*—Merten's experiment with chloroform is described under experiments with alcohol. Herter and Williams gave chloroform by inhalation to 4 dogs. The experiments lasted from six weeks to eight months. They reported marked cirrhotic changes, but gave no precise histologic details and no illustrations of the changes seen.

(Fiessinger intoxicated rabbits with chloroform by injection after the method of Mertens. He studied the stages of injury, degeneration, regeneration and repair which led to cirrhosis.) He found that livers injured by chloroform, toluylene diamine and other substances showed two phases of reaction: (1) a parenchymatous phase evidenced by degeneration and necrosis followed by regeneration and (2) a later phase of fibrous tissue proliferation leading to scarring.

Whipple and his associates studied the effects of diet on the regeneration of hepatic cells following chloroform poisoning. They found that a diet of carbohydrates and milk was most favorable for repair of the liver following chemical injury. Gelatin was found as effective as casein in facilitating repair following a unit injury by chloroform.

Opie and Alvord found that injurious effects of chloroform and of phosphorus on the liver were greater in dogs on a diet of meat than in those on a mixed diet. Also chloroform intoxication produced more extensive necrosis of the liver and more serious systemic disturbances if the animals were given a diet rich in fats. The effect of chloroform was minimized by a diet high in carbohydrates. These observations

have been substantiated by those of Mann on the influence of diet in experimental cirrhosis produced with carbon tetrachloride.

) (Jaffé produced hepatic damage resulting in what he considered cirrhosis by subcutaneous injections of chloroform and of amyl alcohol in rabbits.)

(Schultz, Hall and Baker injected chloroform directly into the portal vein in a series of 80 dogs. This produced immediate necrosis of hepatic cells in the periphery of the lobules.) Frequently extensive groups of lobules were destroyed. Very prompt mobilization of phagocytes occurred. These appeared to be engaged in removing the débris. Simultaneously fibroblastic proliferation occurred. (The areas of necrosis were repaired by regeneration of hepatic cells accompanied by proliferation of bile ducts and by fibrosis. Following such repair the livers were contracted, firm, tough and nodular, and cut with increased resistance. There were heavy bands of connective tissue. Marked cirrhosis was produced, which they stated resembled true (Laënnec's) cirrhosis only superficially.)

(Macchiarulo reported on the effects of chloroform and of chloroform plus cholesterol given to rabbits. Marked parenchymatous and fatty degeneration followed by periportal fibrosis resulted.) He found that chloroform plus cholesterol was more effective in producing hepatic damage than chloroform alone. No illustrations are given.

) (Tar.) Murayama reported cirrhotic changes in the livers of rabbits treated with tar for the experimental production of skin cancer.) Later he reported on the injection of tar and of tar plus hydrous wool fat and of the latter alone into the peritoneal cavities of rabbits. Marked hepatic changes which grossly resembled granular cirrhosis were found in the animals given injections of tar and of tar plus hydrous wool fat. These changes were not seen in those receiving hydrous wool fat alone. The fibrotic changes were such as to cause shrinkage of the liver. They were described as perilobular. No illustrations accompany the report.

(Schirokogeroff noted marked cirrhotic changes in the livers of rabbits whose ears had been painted with tar for the experimental production of cancer.) At his suggestion, Leitman investigated these changes by further experimentation. He painted the ears of 10 rabbits with a mixture of petroleum tar and toluene. These treatments were repeated at intervals of two days. After from two to three weeks the animals became anemic and lost weight. The experiments lasted from one to four months. Marked hepatic changes resulted in each of the 10 rabbits. In the experiments of shorter duration the liver was mottled, firm and finely granular, and cut with resistance. Microscopically it showed marked degeneration accompanied by connective tissue prolifer-

ation. In experiments of longer duration the livers were coarsely granular and contracted. A photograph of the liver presents the hob-nail appearance of Laënnec's cirrhosis (fig. 4A). Photomicrographs show marked cirrhotic changes with evident distortion or destruction

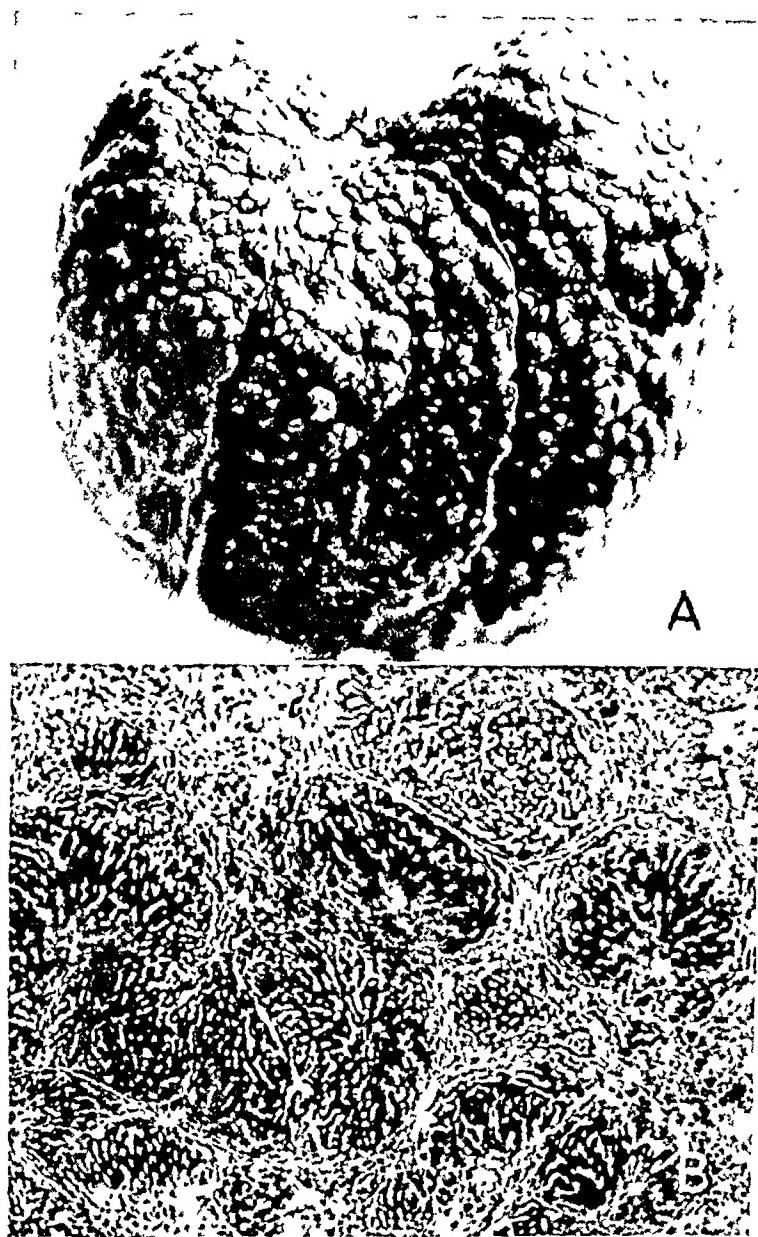


Fig. 4.—*A*, liver from rabbit treated with tar (Leitman). This liver presented both the gross and the microscopic characteristics of portal cirrhosis. *B*, photomicrograph of liver from rabbit treated with tar (Domagk). This shows marked distortion of the lobular pattern. "There are no longer lobules but only nodules of liver cells."

of the lobular pattern and with irregular areas of intercellular fibrosis. In 9 of the 10 the spleen was markedly enlarged and showed hyperplasia of the reticulo-endothelial tissue and hypoplasia of follicles.

Ascites was reported in 7 cases. Rabbits treated with toluene alone showed no such changes.

(Davidson applied coal tar (pix-liquida) to the ears of rabbits at intervals of a few days.) Acute illness with marked loss of weight followed, ending in death within nineteen days. The condition of the liver resembled acute yellow atrophy. Extensive areas of necrosis and marked cellular degeneration accompanied by polymorphonuclear infiltration were present. Chronic effects were seen in rabbits which lived from two to five months following treatment. Abdominal ascites was present, and the liver was rough and nodular. Microscopically, proliferation of fibrous tissue surrounded areas of hepatic cells which were in varying degrees of degeneration and necrosis. There was proliferation of bile ducts with evidences of regeneration of hepatic cells. He believed that acute yellow atrophy, subacute yellow atrophy and cirrhosis are different stages in the same process. Photographs of gross specimens and of microscopic cross-sections substantiate the author's description. (Distortion of lobular pattern characteristic of Laënnec's cirrhosis is shown.)

(Domagk applied tar to the ears of rabbits in attempts to produce cancer. The animals became cachectic. The livers were indurated and had roughened granular surfaces.) There was a marked increase in fibrous tissue enclosing nodules of hepatic cells. His descriptions and photographs show marked distortion, if not obliteration, of the lobular pattern (fig. 4B). The condition closely resembled Laënnec's cirrhosis as seen in man. These results were found in each of 6 rabbits following the application of tar from fifty-seven to ninety-five times. In each instance there was atrophy of splenic pulp; ascites was reported in 4 of the 6 rabbits.

(Polson gave shale oil in doses of from 2 to 10 cc. to rabbits subcutaneously, intraperitoneally, intrapleurally and *per vaginam*. Single large doses produced extensive hepatic necrosis. Hepatic damage in all stages from necrosis to cirrhosis resulted from repeated smaller doses. Cirrhosis having all the features of atrophic cirrhosis in man was reported.) Each experiment was controlled by a biopsy on a specimen of the liver obtained by laparotomy before treatment of the animal. Photographs show a rough nodular condition of the liver resembling Laënnec's cirrhosis. One of the photomicrographs shows distortion of the lobular pattern.

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912 (c) *Carbon Tetrachloride and Related Substances*.—Fiessinger, Wolf and Blum subjected mice to an atmosphere charged with tetrachlorethane by evaporation for one and a half hours repeatedly.) This produced stupor and other symptoms resembling those from alcohol or ether. The mice became jaundiced, the urine contained bile, and at autopsy the tissues were visibly icteric. (The livers had a nutmeg-grater

appearance.) After from three to four days nuclear and cytoplasmic changes were seen. The mitochondria became enlarged and coarse. Later they were transformed into fatty droplets, and the cytoplasm became homogeneous. (Repair began about the seventh day. Regeneration of hepatic cells was followed by connective tissue proliferation and cicatrization in the periphery of the lobules.) The authors emphasized this sequence in all toxic injuries of the liver.

(Ottenberg and Abramson gave tetrachlorophenolphthalein and tetrabromophenolphthalein to dogs and rabbits in doses of from 0.3 to 0.4 Gm. per kilogram. This produced violent illness, evidenced by weakness, staggering, vomiting and frequent defecation. Death resulted within eight hours. Examination showed extensive necrosis, hemorrhage and disruption of the hepatic architecture. Chronic effects were not studied.

Gardner and his associates gave carbon tetrachloride to dogs and rabbits by mouth, by inhalation, by subcutaneous and intraperitoneal injections and per rectum. In every instance extensive central necrosis of the liver resulted. There were no other lesions of consequence. Regeneration and healing began three or four days after the intoxication. They found that the addition of alcohol to carbon tetrachloride greatly increased its toxicity. Healing of the degenerated areas was accompanied by marked central fibrosis. Their photomicrographs confirm the observations but do not show obliteration of the lobular pattern.

Midorikawa gave 0.05 cc. of carbon tetrachloride to rabbits by subcutaneous injection at intervals of from five to seven days. This resulted in pronounced parenchymatous degeneration, hydropic degeneration and fatty changes in the liver. Congestion, stasis and hemorrhages were also present. These changes were followed by round cell infiltration and by proliferation of hepatic cells, bile ducts and connective tissue. The formation of pseudolobules was reported. No illustrations accompanied this article.

(Lamson and Wing gave carbon tetrachloride and alcohol, separately and in combination, to three series of dogs. The experiments extended from five to six months. The livers were greatly scarred, granular and contracted following chronic intoxication with carbon tetrachloride. The combination of carbon tetrachloride with alcohol was more effective than carbon tetrachloride alone.) Alcohol alone produced no visible changes in the livers. No changes were seen in other organs. Microscopic examination showed distinct scarring and distortion of lobular architecture. The photomicrographs indicate that the lobular pattern was obliterated.

(Albot produced cirrhosis in rats, guinea-pigs and rabbits by giving carbon tetrachloride by inhalation.) After two hundred and thirty days there was marked proliferation of fibrous tissue with atrophy of hepatic cells resulting in marked distortion of architectural pattern.

Van der Schueren gave 0.25 cc. of carbon tetrachloride twice weekly to rabbits and guinea-pigs. Extensive cirrhosis resulted from four weeks' treatment. This was described as having the characteristics of Laënnec's cirrhosis, and led the author to conclude that degeneration and destruction of parenchyma are primary, and that regeneration and proliferation are secondary processes.

Mann, Bollman, Anderson, Fishback, Higgins, Lacquet, Love, Priestly, Stephenson and Williamson have experimented by means of successive removals of portions of the liver, chemical injuries, dietary influences, circulatory conditions and other factors affecting the liver. The chief purpose of these studies was physiologic, but their bearing on cirrhosis is important. Following the work of Gardner, Lamson and Wing and others, they adopted carbon tetrachloride as a toxic agent producing permanent damage to the liver more accurately and effectively than either phosphorus or chloroform.

(To dogs of average size Bollman and Mann gave from 3 to 5 cc. of carbon tetrachloride by stomach tube twice weekly. Such doses produced necrosis of liver cells followed by regeneration and fibrosis.) Following a single toxic dose it was found that the reparative processes began within twenty-four hours. There was marked infiltration in which polymorphonuclear cells predominated at first, and mononuclear cells later. Disintegrated hepatic cells were removed, in part, by phagocytosis and were replaced by multiplication of adjacent uninjured cells. Proliferation of fibrous tissue apparently occurred from that present about the portal spaces. Repetition of the injury gave rise to extensive scarring of the liver; hypertrophic nodules developed forming islands of hepatic cells without normal relationship to the blood vessels and to the biliary ducts. The lobular pattern became distorted as in typical Laënnec's cirrhosis. After marked cirrhosis had been established it continued to progress slowly without further administration of the poison. The gross and microscopic appearances of the liver showed close resemblance to Laënnec's cirrhosis. The liver was very nodular and granular, and the microscopic picture showed alteration of architecture with distortion of the lobular pattern.

Vascular changes resembling those of Laënnec's cirrhosis developed. The total blood supply to the liver was diminished, owing to the cicatricial constriction about the vascular terminals. Varices and collateral circulatory channels were formed. Hemorrhage occurred in the stomach and bowel, and voluminous ascites developed. Quantities of from 1,000 to 3,000 cc. were removed repeatedly from some of the animals by tapping. It was found that diet had a marked influence on the development of ascites. Little or no ascites formed if the dogs were fed with

syrup and other carbohydrates. When a high protein diet was substituted, ascites became marked. This was again absorbed when the dogs were given carbohydrates. This evidence corroborates the observations of Opie, Whipple and their associates on the influence of diet on hepatic repair. These findings led Mann and his co-workers to conclude that, at least in their animals, the formation of ascites is not conditioned entirely by obstruction to the portal blood flow.

They were unable to produce necrosis of hepatic cells with alcohol alone, but a combination of alcohol and carbon tetrachloride was more toxic than carbon tetrachloride alone. They found that the toxic agents were more effective when given to fasting than when given to well nourished animals. The exclusion of the portal blood from the liver by means of an Eck fistula rendered the liver ten times more susceptible to carbon tetrachloride than is the normal liver. Eck's fistula also decreased the regenerative capacity of the liver. Surgical removal of a portion of the normal liver was followed by prompt restoration of the liver to its normal volume. In dogs with Eck fistula such removal was not followed by restoration. Apparently the normal flow of portal blood through the liver aids in its resistance to injurious agents and its regeneration following injury. In human cirrhosis interference with portal blood flow probably renders the liver more susceptible to injury and less capable of regeneration.

When well advanced cirrhosis had been established by treatment with carbon tetrachloride, portions of the involved livers were removed surgically. This was followed by very deficient restoration as compared with that following similar operations on normal dogs.

Ligation of the common bile duct was performed on a series of dogs. This produced marked jaundice, which decreased after a few weeks. The cells of the parenchyma showed little, if any, atrophic or other change, but those bordering the bile ducts were deeply pigmented, and there was evident proliferation of bile ducts with slight increase of connective tissue about them. Infection played an important part in these experiments. When infection occurred following ligation of the duct, the formation of connective tissue was marked, and there was atrophy of the parenchymal cells. In such experiments when one or more lobes of the liver were removed surgically, the regenerative capacity was found to be greatly lowered.

Mann and his collaborators also found that the transplantation of the common bile duct into any portion of the gastro-intestinal tract or the anastomosis of the gallbladder to any portion of the gastro-intestinal tract was invariably followed by severe effects in the liver. The changes varied from moderate cholangitis to marked cirrhosis. The type of cirrhosis is not specified but it is assumed that it was biliary.

Mann emphasized three factors tending to prevent the restoration of the cirrhotic liver in experimental animals: extensive cicatrization of the liver, reduction of the blood supply and jaundice.

(d) *Miscellaneous Organic Substances.*—Boix produced degenerative changes followed by necrosis and proliferation of fibrous tissue and bile ducts, resulting in what he regarded as cirrhosis. These changes followed the use of butyric acid, valerianic acid and acetic acid.) His work is frequently quoted as significant since these are products which might result from gastro-intestinal dysfunction. If such dysfunction with the production of such acids were an important factor in the development of cirrhosis, it would seem that a higher percentage of cirrhosis should be found among those suffering from various gastro-intestinal disorders. De Josselin de Jong, D'Amato and others failed to produce cirrhosis by feeding butyric acid.

Poggendorf mixed butyric acid with the food given to 6 rabbits over periods varying from thirty-two to one hundred and sixty-three days. He described a perilobular inflammation with a lymphocytic infiltration and a proliferation of connective tissue which he interpreted as the initial stage of interlobular cirrhosis. Chronic inflammation of the pancreas and catarrhal gastro-enteritis also were reported. The results are illustrated by drawings which indicate moderate periportal inflammation.

Inghilleri mixed fecal material from patients with hyperchlorhydria with the feed of rabbits. He reported chronic inflammation of the liver showing proliferation of connective tissue and of bile ducts. In another series he mixed the food with intestinal contents obtained from patients with hyperchlorhydria. This mixture was incubated forty-eight hours and fed to animals. Evidences of hepatic damage were reported as more marked in the latter than in the first group.

Chalatow mixed cholesterol with food given to 8 rabbits during periods of from fourteen days to four and one-half months. He found a pronounced deposit of anisotropic fats in the livers. Several of the livers showed an increase in fibrous tissue with cellular infiltration, and in one liver the condition resembled granular cirrhosis grossly and microscopically.

Hayami produced hepatic damage resulting in necrosis by injecting aleuronat into the portal veins. Healing of these lesions resulted in irregular scarring of the liver.

Adler mixed tobacco infusion with food given to rabbits. This produced interlobular accumulations of round cells followed in cases of long duration by a proliferation of fibrous tissue which occasionally penetrated slightly into the lobules. There was no distortion of the lobular pattern. Ogata injected an aqueous extract of tobacco leaves

into the ear veins of rabbits. Slight degenerative changes of the liver were seen but no fibrosis.

Rovighi and Portioli produced degeneration and leukocytic infiltration in the livers of rabbits by feeding them ammonium carbamate. Joannovicz confirmed these experiments with ammonium carbamate. He produced similar changes with toluylene diamine in dogs. This resulted in degeneration followed by proliferation of interlobular connective tissue and of bile ducts. Icterus was observed in some of the animals.

Hensen injected such poisons as chloroform, formaldehyde and sulphuric acid into the livers of cats by way of the bile ducts. Chloroform produced fatty changes and necrosis followed by restitution. Formaldehyde and sulphuric acid produced extensive necrosis followed by gradual resorption and regeneration. The regeneration and healing were accompanied by proliferation of connective tissue.

Krawkow mixed an infusion of putrid horse flesh with the food given to dogs for several weeks. He reported genuine cirrhosis following these experiments. He also injected cultures of *Bacillus pyocyaneus* into doves and observed cirrhotic changes in their livers.

Ignatowski fed milk, meat and eggs to young rabbits, and reported that the livers became cirrhotic. Ascites was present in 1 animal. His description indicates marked degenerative changes of the liver with periportal fibrosis. The spleen was regularly enlarged, and atheroma of the aorta and parenchymatous degeneration of the kidneys were reported. The drawings show periportal inflammation and proliferation of connective tissue, without distortion of lobular pattern.

Flexner produced intoxications of guinea-pigs and rabbits with bacterial protein, ricin, abrin and foreign serums, and found necrosis of hepatic cells followed by leukocytic infiltration fibrosis and proliferation of bile ducts in animals that survived several weeks. He stated that grossly the livers were firm and somewhat granular, but that they did not reproduce the gross or the microscopic features of human cirrhosis.

Deutsch prepared an antihepatic serum by immunizing rabbits against the hepatic substance of other animals. The injection of such serum into the peritoneal cavities of animals produced necrosis of the liver. Cantacuzene reported similar results. Delezenne injected antihepatic serum into the peritoneal cavities of dogs. The dosage was from 2 to 4 cc. per kilogram of body weight. This produced severe intoxication and death in from fifteen to twenty-four hours. The condition of the livers resembled acute yellow atrophy. In animals which survived from five to fifteen days there was marked inflammatory reaction with leukocytic infiltration and proliferation of fibrous tissue. This was accompanied by regeneration of liver cells. No illustrations accompany the

article. Joannovcz reported degenerative changes in the liver followed by regeneration and proliferation of fibrous tissue, resulting from injections of immune serums.

Fukuhara produced intoxication of rabbits, guinea-pigs, rats, dogs and hens by injecting foreign proteins, bile and hemolytic serums. Localized necroses of hepatic cells were found. Parenchymatous changes were seen in other tissue. The chronic effects of such agents were not studied.

Civray found that the injection of specific hepatolytic serum into animals caused acute degeneration, necrosis and cytolysis of hepatic cells. Other organs were affected in minor degrees. Chronic or repeated effects of such treatment were not studied.

Pearce gave dogs injections of serums from rabbits which had been immunized to dog's red blood corpuscles. Some of the dogs so treated died in from four minutes to forty-eight hours. In these there was diffuse necrosis of the liver. In the livers of those which lived forty-eight hours or longer, proliferation of endothelial cells and of connective tissue cells was seen. In five days there was marked regeneration of hepatic cells with proliferation of granulation tissue. In 1 dog thirty-six days after injection of the serum the liver was firm and had a finely granular surface. On section, distinct pseudolobulations were seen. Histologically there was marked perilobular fibrosis with lymphoid infiltration and proliferation of bile ducts. The reparative process was most active in the region of the portal spaces and resulted in diffuse cirrhosis. Illustrations show marked cirrhosis but no distortion of lobular architecture. Pearce did not claim that this experiment explained the origin of cirrhosis in man, but that it proved that cirrhosis results from a reparative process following parenchymatous injury.

Jaffé reported perilobular fibrosis in rabbits following repeated injections of a 10 per cent solution of hemoglobin.

Wells produced chronic perilobular inflammation which he described as cirrhosis, by subcutaneous injection of commercial peptone into guinea-pigs and rabbits. The livers were indurated, the fibrous tissue proliferation did not penetrate the lobules, and there was no distortion of the lobular pattern.

D'Amato gave rabbits and dogs injections of meat juices and broth. He reported marked congestion of the liver in each instance. In some there were degeneration and necrosis associated with round cell infiltration about the portal structures.

Longcope produced anaphylactic shock in dogs, cats, rabbits and guinea-pigs which had been appropriately sensitized to egg or serum albumin. Repeated anaphylactic intoxication resulted in necroses of groups of cells in the periphery of the hepatic lobules. The healing of

these resulted in a proliferation of scar tissue which was perilobular in location. There was no distortion of the lobular architecture.

Lissauer injected a sterile extract of horse flesh intravenously into 4 rabbits. Doses of 5 cc. of the extract were given at intervals of six days. The experiments lasted from thirty-four to seventy days. His descriptions and drawings indicate moderate perilobular hepatitis.

Apparently several compounds of the benzine series have the property of producing damage to the parenchyma of the liver. Iwanoff found marked parenchymatous and fatty degeneration of the hepatic cells following treatment with antipyrine. No fibrous tissue proliferation was reported. Marckwald produced local necroses of the liver by subcutaneous injection of antipyrine into frogs and rabbits. A marked perilobular fibrosis with proliferation of bile ducts resulted in what he regarded as a typical picture of cirrhosis.

Some significant observations concerning the effects of aniline, toluidine and methenamine have resulted from experiments not primarily designed to produce hepatic damage or cirrhosis. It had been noticed that those who work with aniline compounds frequently acquire primary neoplastic growths. Jaffé undertook an experimental investigation of this phenomenon. Groups consisting of equal numbers of rabbits, guinea-pigs and mice were given aniline, toluidine and methenamine in gaseous form by inhalation. The average duration of the experiments was six months; the longest duration was one year and four months. No neoplastic growths resulted in any of the animals, and no significant changes were seen except in the livers. The animals treated with aniline and those with toluidine showed marked parenchymatous and fatty changes of the liver. Those receiving methenamine presented necroses accompanied by regeneration of liver cells, round cell infiltration and proliferation of connective tissue. Jaffé saw in these results a relationship to the experimental production of cirrhosis.

In another experiment he reported cirrhosis in rabbits following injections of hydrazine sulphate and of phenol hydrazine hydrochloride. Large doses of these agents produced extensive necrosis of hepatic cells. Smaller doses repeated over long periods produced degeneration followed by marked connective tissue proliferation and lymphocytic infiltration. His illustrations confirm these findings, but they show no distortion of the lobular architecture.

Hurst and Hurst injected phenylhydrazine in doses of from 0.1 to 0.17 Gm. into rabbits and guinea-pigs. Some necrosis of hepatic cells followed acute intoxication with phenylhydrazine, but no distinct cirrhosis resulted from chronic intoxication with it. The injection of quantities of sheep bile or human bile into animals was not followed

by degeneration and fibrosis of the liver. Injections of guanidine likewise resulted in no hepatic structural changes.

*Cinchophen.*—Sherwood and Sherwood collected from the literature 48 instances of cinchophen poisoning in man, 15 of which ended fatally. It was noted that extensive hepatic necrosis is characteristic of cinchophen poisoning.

Churchill and Van Wagoner fed cinchophen daily to dogs in a dosage of 595 mg. per kilogram of body weight. The dogs died in from ten to twenty days. The livers showed areas of necrosis of varying extent. Prolonged treatment with cinchophen produced gastric and duodenal ulcers, but cirrhosis was not seen.

Reichle gave cinchophen subcutaneously to rats. A single dose of 1 Gm. per kilogram of body weight caused death within twenty-four hours. The liver, kidneys and spleen showed extreme degeneration and necrosis of cells. Doses of from 0.2 to 0.5 Gm. per kilogram of body weight were given daily for eight days. Marked illness and partial paralysis followed. The organs showed severe parenchymatous degeneration and focal necrosis. When the treatment was discontinued recovery followed. Seventeen days later the liver had returned to a normal histologic picture. The healing process was not accompanied by fibrosis. Similar results were obtained in rats when their livers had been depleted of glycogen by starvation.

Barbour and Fisk gave cinchophen, neocinchophen and sodium salicylate by mouth to groups of dogs and rats. Acute hepatic degeneration and necrosis resulted from the activity of each. They found that the order of hepatic toxicity from these drugs was cinchophen > sodium salicylate > neocinchophen. No cirrhotic changes following cinchophen were found in six weeks. Gastric or duodenal ulcers were found in 4 of 8 dogs receiving cinchophen; none were found following sodium salicylate or neocinchophen.

Beaver and Robertson, also Permar and Goering reported on cinchophen poisoning in man. They confirmed previous observations that cinchophen produces extensive necrosis resembling that of acute yellow atrophy. But important differences in the two conditions were emphasized. Cinchophen produces a form of atrophy of the liver in which there is marked retardation of regeneration and in which proliferation of fibrous tissue and of bile ducts is not seen.

The experimental studies and the postmortem observations in man indicate that cinchophen produces marked hepatic damage, but that this may lead to cirrhosis has not been shown.

Fischer reported "typical cirrhosis of the liver" resulting from the intravenous injection of ether in "leinol" into mice and rabbits. His report contains no histologic details and no photographs.

## (INFECTIOUS AGENTS)

The belief that infections are important factors in the causation of cirrhosis has become rather general among pathologists. Mallory assigned infection as the cause for one group of cirrhosis; Beattie and Dickson stated that cirrhosis may occur secondary to infectious disease and regarded scarlet fever as of importance, especially in young subjects. Muir mentioned infection, especially scarlet fever, as important and stated that a fair proportion of cases are postinfective. Karsner included infection among the probable etiologic agents. (Boyd believed that the major factor is probably a low grade bacterial infection.) De Josselin de Jong found numerous authors in agreement that long-continued toxic or infectious processes were of primary importance. Fiessinger believed that infections are of major importance in tropical and subtropical countries. Yenikomshian found that cirrhosis is particularly prevalent in the rural population of Tyre and Sidon where alcoholic drink is taboo. A history of amebiasis or malaria was regular in such cases, and he believed that a combination of the two is the most important cause for cirrhosis in that population.

Wolff injected putrefactive bacteria subcutaneously into animals. This was followed by an interstitial hepatitis with round cell infiltration and connective tissue proliferation. In some of the animals this process was diffuse; in others it was localized and resulted in nodular lesions.

Roger found that experimental infections with streptococci or anthrax bacilli lowered the glycogen content of the liver and increased its susceptibility to injury. He produced interstitial hepatitis by repeated injections of "Bacillus septicus putridus."

Scagliosi produced beginning cirrhosis by repeated inoculation of nonpathogenic bacteria into alcohol-treated animals. Rabbits, guinea-pigs and chickens were used. He believed that alcohol acts as a "near cause" in producing hepatitis, a view which has found support among pathologists in recent years.

Dantschakow-Grigorevski injected broth cultures of staphylococci of low virulence into rabbits during periods of from seven to fifteen weeks. The number of animals was not stated. The livers of 2 were described as large and firm. Drawings illustrate periportal inflammation.

Brieger reported cirrhosis in rabbits and guinea-pigs following inoculation of tubercle bacilli. Hanot and Gilbert reported the production of cirrhosis following injections of tubercle bacilli of lowered virulence. Besançon, Griffon and others reported similar results from experiments with *Bacillus tuberculosis*. Joanovicz described the production of cirrhosis in guinea-pigs following the injection of urine from tuberculous kidneys. Ogata injected a sterilized suspension of tubercle bacilli into the portal veins of rabbits. This resulted in "tuberculous cirrhotic

changes." A sterile suspension of colon bacilli similarly injected produced slight inflammatory changes in the liver.

(Adami investigated an epidemic disease of cattle which produced marked cirrhosis resembling that seen in man.) He found in the livers numerous coccoid organisms which, when cultivated, resembled the bacillus of hemorrhagic septicemia. When inoculated into rabbits the organisms produced marked degeneration and necrosis of the liver. The animals died early before cirrhotic changes could occur. (Adami regarded "sub-infection" as important in producing cirrhosis)

At this point attention is called to the frequent occurrence of cirrhosis in rabbits as a result of chronic coccidiosis and to its occasional occurrence in guinea-pigs as a result of an infection resembling tuberculosis. If spontaneous infections produce cirrhosis in animals it is probable that they may do so in man.

Flexner, whose work is referred to in another section, included bacterial substances among the injurious agents that produced cirrhotic changes in guinea-pigs and rabbits.

Kirikow cultivated diplococci from the liver in 4 cases of what he regarded as Hanot's cirrhosis. In each case the condition was acute and was accompanied by fever, icterus and enlargement of the liver and spleen. The liver was described as distinctly granular, containing nodules of hyperplastic cells. The organisms isolated were pathogenic for mice. Those which survived the initial inoculation were reinoculated repeatedly. This produced marked diffuse hepatitis, with inflammatory infiltration and fibrosis of the liver, accompanied by splenic enlargement.

Weaver reported the isolation from a spontaneously infected guinea-pig of a bacillus not definitely identified but belonging to the colon group. When inoculated into guinea-pigs this organism produced marked hepatic necrosis followed by death. Animals that lived seven days or longer presented cirrhotic changes. These consisted of regenerated liver cells and proliferation of bile ducts and of fibrous tissue. No illustrations are given. This organism became avirulent in artificial mediums.

Hektoen isolated from human blastomycotic lesions an organism which morphologically resembled *Bacillus diphtheriae* but differed from it in certain particulars. When cultures of the organism were inoculated into guinea-pigs, rabbits and dogs, it produced early death with marked degeneration, necrosis and acute inflammation of the liver. In animals that survived two weeks or more the liver showed inflammatory changes with perilobular fibrosis and proliferation of bile ducts. These cirrhotic changes were described as portal. Photomicrographs show marked cirrhosis with penetration of connective tissue into the lobules and with some distortion of the lobular pattern. The inoculated organism was recovered in subculture from the livers of some of the animals.

The organism became avirulent in later cultures, and subsequent experiments were without results.

(Opie's experiments included combinations of toxic degeneration of the liver with bacterial infections.) In one series he gave chloroform in amounts of from 0.5 to 1 cc. per kilogram to dogs at intervals. This was followed by marked degeneration and destruction of hepatic cells in the central portion of the lobules. Regeneration of the cells and proliferation of connective tissue and of bile ducts followed. In dogs that lived from two to three months, marked cirrhosis, jaundice and dilatation of the portal vein resulted. The fibrosis began about the central vein and extended into the lobules. Following repeated injury of this kind distortion of the lobular architecture resulted. Opie combined chloroform intoxication with intravenous injections of streptococci and colon bacilli. In animals examined at an early date this produced a picture resembling acute yellow atrophy. In animals that lived longer (twenty-three days) there was marked cirrhosis accompanied by proliferation of bile ducts. Photomicrographs show marked distortion of the architecture in some; in others the lobular pattern was visible. No ascites was reported in these animals.

Evidence indicating a relationship between infection and cirrhosis has been drawn from clinical observations and postmortem studies. In some instances such evidence has a significance second only to that of well controlled experimentation. A number of reports have indicated that streptococcal infections are related to the development of cirrhosis. Many of these were made before streptococci were known to be related to scarlet fever and measles.

Siredey described degenerative and inflammatory changes in the liver following diphtheria and also scarlet fever. He believed that such inflammation may become chronic and may account for the development of cirrhotic changes. He regarded alcoholism as a contributory factor making the patient more susceptible to infection.

In certain cases of scarlet fever and of measles Henoch found chronic lesions which constituted an interstitial hepatitis. He described marked degenerative changes associated with proliferation of bile ducts and of connective tissue. He regarded this as a form of hepatitis that might develop into cirrhosis.

Mogk reported the case of a child whose death from cirrhosis occurred eight weeks after a severe attack of scarlet fever. Streptococci were demonstrated in sections of the liver. The active process resembled those later described by Moon in similar cases.

Gastou described inflammation about the portal structures associated with diffuse hepatitis following acute infections. In such a liver from a child he found streptococci in chains. In 2 cases of cirrhosis, he

cultivated diplococci from the blood and from the hepatic substance. He believed various organisms caused infectious hepatitis which later produced cirrhosis.

Babes reported 4 cases of streptococcic septicemia which resulted in widespread degeneration and necrosis in the liver. Streptococci were cultured from the blood in each case, and similar organisms were found in microscopic examination of the liver. In 3 cases the condition of the liver resembled acute yellow atrophy.

Levi reported cirrhosis associated with mitral endocarditis in a boy 17 years of age. He cultivated from the liver a diplococcus which was pathogenic for guinea-pigs.

Bingel studied the liver in 8 cases of delayed death following scarlet fever. Extreme degeneration, necrosis of hepatic cells and perivasculär inflammatory infiltration were found in every case. In the liver of one patient whose death occurred seven months after scarlet fever an advanced stage of cirrhosis was found. Bingel believed the cirrhosis in such cases to be a sequel of the infection.

Baginsky reported a case of streptococcic septicemia in a child. The liver showed interstitial hepatitis characterized by extensive degeneration and necrosis of hepatic cells and by a rather diffuse inflammatory reaction. Streptococci were cultivated from the blood stream and from hepatic substance obtained by puncture prior to death.

Rolleston and McNee included streptococci among the agents which may produce icterus gravis. It is generally believed that cirrhosis may develop from icterus gravis or from recurring attacks of toxic jaundice in which the etiology may be similar.

The occurrence of portal cirrhosis in children is not unusual, and it occasionally affects more than one in the same family. In such instances some acute infectious disease usually precedes. Howard reported 2 such cases of cirrhosis in a brother and sister aged 8 and 9 years. The disease developed following pertussis, and he included infection among the probable causes. Ely reported cirrhosis in twins 4 years old. The disease began with jaundice during an attack of measles and ended in death five months later. Moon reported 3 cases of cirrhosis in one family. Scarlet fever preceded the development of the cirrhosis. In 1 case hemolytic streptococci were cultivated from the substance of the liver at postmortem examination. A blood culture made a few hours prior to death was negative. This indicated that the streptococci found in the liver were not incidental to a terminal septicemia. The streptococci were inoculated into young rabbits by various routes. The animals lived from three to fifteen days, and in 11 of 12 rabbits the hemolytic streptococci were recultivated from the livers. Marked degeneration and necrosis and very little inflammatory reaction were present in the livers.

( Moon and his associates cultivated streptococci from cirrhotic livers in 6 cases, and demonstrated cocci in sections in 9 others in which the cirrhotic process was active. Failure to cultivate or demonstrate organisms in a number of similar cases indicated that all were not due to the same cause.)

( MacMahon and Mallory reported 5 cases of streptococcal hepatitis, in 4 of which they demonstrated streptococci in the liver.). Two were acute and showed degeneration and necrosis of hepatic cells with an associated inflammatory reaction. One was subacute, of six weeks' duration, and showed marked zonal necrosis affecting all the lobules. There was proliferation of hepatic cells and of fibrous tissue such as is characteristic of early cirrhosis. In another case the liver, seen during cholecystotomy, appeared normal, and an excised section had normal histologic features. Subsequently chills and fever developed, accompanied by jaundice. The liver became enlarged; ascites developed and was removed by tapping on two occasions. The condition progressed to a fatal termination eight and one-half months following the operation. Pronounced cirrhosis was found post mortem, and numerous streptococci were demonstrated in the liver. They regarded this as a chronic hepatitis due to streptococci of moderate virulence. This has caused a diffuse progressive destruction of hepatic cells resulting in cirrhosis. Guinea-pigs and rabbits, inoculated via the portal vein with *Streptococcus scarlatinae*, showed localized and diffuse areas of hepatic necrosis. Chronic effects were not studied. ( They believed that streptococci may cause hepatic necrosis resembling that of acute yellow atrophy, and that it may develop into cirrhosis.)

( In view of the fact that infections outrank all other agents in the production of chronic inflammatory processes, it is strange that they have received so little investigation as possible causes of cirrhosis.)

#### OBSTRUCTION TO VESSELS AND DUCTS

Obstructions to the hepatic vessels have been produced experimentally by various means and in various combinations. These have resulted in circulatory, degenerative and atrophic changes in the parenchyma of the liver. No condition resembling portal cirrhosis has been reported from such experiments. Experimental obstruction to the hepatic duct produces pathologic conditions which vary somewhat depending on the duration and the degree of the obstruction and on the presence or absence of associated infection. Such obstructions have not produced portal cirrhosis, though the distinctive structural changes of biliary cirrhosis have been reproduced satisfactorily by such means.

No review of such experiments will be attempted since there is general agreement as to the origin and development of biliary cirrhosis.

Occasionally such a condition becomes progressive and the parenchymal injury is no longer limited to the periphery of the lobules. In such an instance extensive injury and repair may lead to obliteration of the lobular pattern and to obstruction of portal circulation. Thus in rare cases the characteristic features of portal cirrhosis may be superimposed on those of biliary cirrhosis.

#### SUMMARY

A review of experimental cirrhosis shows that numerous agents, most diverse in character, are capable of causing degeneration and necrosis of hepatic cells. These include various chemicals, both organic and inorganic, certain drugs and tarlike substances, foreign proteins and products of protein decomposition, bacterial products, immune serums and infections. The continued or repeated action of these agents has resulted in the production of chronic diffuse hepatitis, as indicated by degeneration and necrosis, by regeneration of parenchyma and by proliferation of fibrous tissue.

Probably the majority of these agents have no etiologic significance in the development of human cirrhosis. Yet the results of the investigations of their action have contributed greatly to an understanding of the mechanism by which cirrhosis develops. In every instance in which the prolonged action of an agent has resulted in some degree of cirrhosis the acute effects have been degeneration and necrosis of hepatic cells. Cirrhosis, like other chronic inflammations, results from parenchymatous injury followed by repair. If such injury is severe, continued or repeated, the repair will be accompanied by fibrosis. This evidence should tend to allay a controversy of long duration between certain groups of European pathologists.

A few agents which have met satisfactorily the rigid criteria for Laënnec's cirrhosis are phosphorus plus alcohol, manganese chloride plus phenylhydrazine, carbon tetrachloride, tars, bacterial infections and combinations of infection with other agents, as in Opie's experiments.<sup>19,20</sup> Each of these has produced a distortion of the architecture of the organ, in which marked alteration of lobular pattern was evident. The islands of regenerated hepatic cells occurred as nodules rather than lobules, and indicated the type of circulatory disarrangement which is characteristic of Laënnec's cirrhosis. In Mann's experiments<sup>19,21</sup> with carbon tetrachloride the circulatory disturbances resulted in portal circulatory obstruction, as indicated by ascites and by the development of collateral venous channels.

(The belief that cirrhosis is caused by alcohol has not received experimental support. Without exception the agents which have produced cirrhosis experimentally have caused hepatic necrosis. Alcohol, even in large amounts and with long-continued use, has caused only parenchyma-

tous degeneration and fatty changes. These have not resulted in necrosis nor in permanent hepatic changes. However, the probability that alcohol acts as a contributing or predisposing factor has received experimental support.) Alcohol has been found to accentuate the injurious effects of bacteria, of phosphorus, of chloroform and of carbon tetrachloride on the animal liver. It is probable that alcohol may similarly accentuate the effects of injurious agents on the human liver. By virtue of this property alcohol may be an important contributory factor in the development of human cirrhosis.

Cirrhosis resulting from treatment with tar has duplicated Laënnec's cirrhosis with remarkable accuracy. The gross and microscopic characteristics have fulfilled rigid requirements. The splenic changes have resembled those accompanying Laënnec's cirrhosis, and ascites has been an almost constant associated feature. The fact that the experiments were made on rabbits makes necessary a high degree of conservatism. However, the very high percentage of positive results makes it probable that coccidiosis was not concerned. In Polson's experiments erroneous conclusions were partially obviated by examinations of the livers prior to experimentation.

The injurious effects of hydrazine and similar organic chemicals and the production of cirrhosis by means of tars, have perhaps a deeper significance than is apparent. Many investigators have believed that various substances present incidentally in alcoholic liquors might be responsible for cirrhosis occurring among drinkers of those beverages. Many such contaminants have been investigated without significant results. The tarlike products or substances of the benzine series formed by the charring of casks used for the ageing of liquor have not been investigated. Liquors aged in charred casks contain visible quantities of a brownish substance dissolved from the semicharred wood. Such substances might produce effects similar to those of tars. Is it possible that such substances may be responsible for the development of cirrhosis in habitual drinkers of alcoholic liquors?

There is evidence that metabolic disturbances may be important contributing factors. Medical literature contains record of many observations on the simultaneous occurrence of exophthalmic goiter and cirrhosis (Marine, Goodpasture, Raab and Terplan, Weller, Assmann, Haban, Beaver and Pemberton). Moon's survey on the occurrence of cirrhosis in different sections of the United States showed a most suggestive relationship between cirrhosis and thyroid disease. The states having the highest death rate from thyroid disease had also the highest death rate from cirrhosis and vice versa. A similar relationship is exemplified in European countries. The highest occurrence of cirrhosis on record is in Switzerland, where goiter also is most prevalent. Livers

with low glycogen content have an increased susceptibility to injury. Hyperthyroidism may contribute to the development of cirrhosis by producing hepatic glycopenia. Rössle and others have shown that marked thyrotoxicosis produces extensive hepatic necrosis. Hence thyrotoxicosis may be a direct cause of cirrhosis.

Infections in animals are known to cause a hepatitis which resembles Laënnec's cirrhosis closely. The experiments of Weaver, Hektoen, Opie, Mallory and others have been cited. The high incidence of cirrhosis in tropical countries, associated with infectious diseases peculiar to those regions, is significant. It is admitted that congenital syphilis may result in a form of cirrhosis resembling that bearing the name of Laënnec. Many observers have demonstrated a relationship between streptococcic infection and cirrhosis. It is probable that chronic diffuse infections of the liver are important factors in the etiology of cirrhosis.

It is significant that combinations of agents have been found more effective than either of the same agents alone. The combination of alcohol with phosphorus, of manganese chloride with colon bacilli and of that salt with hydrozine, of chloroform with alcohol, cholesterol, streptococci or colon bacilli, of alcohol with bacteria, and with carbon tetrachloride, and the effects of diet in experiments with phosphorus, chloroform and carbon tetrachloride, are important examples. The results in such instances suggest the importance of combinations of agents in the etiology of human cirrhosis.

A survey of experimental and clinical studies on cirrhosis convinces one that no single causative factor is responsible. Cirrhosis is synonymous with chronic progressive hepatitis. Its etiology will be found to be as variable as are the agents which, singly or in combinations, may cause chronic diffuse progressive inflammation of the liver.

#### BIBLIOGRAPHY

For references to reports prior to 1910, the reader is referred to the following reviews:

- van Heukelom, S.: *Beitr. z. path. Anat. u. z. allg. Path.* **20**:221, 1896.
  - Klopstock, F.: *Virchows Arch. f. path. Anat.* **184**:304, 1906.
  - Fischler, F.: *Deutsches Arch. f. klin. Med.* **93**:427, 1908; *Ergebn. d. inn. Med. u. Kinderh.* **3**:240, 1909.
  - Saltykow, S.: *Zentralbl. f. allg. Path. u. path. Anat.* **22**:849, 1911.
  - Rössle, R., in Henke, F., and Lubarsch, O.: *Handbuch der speziellen pathologischen Anatomie und Histologie*, Berlin, Julius Springer, 1930, vol. 5, p. 489.
- 

Adami, J. G.: *Lancet* **2**:396, 1898; *Brit. M. J.* **2**:1215, 1898.

Adler, I.: *J. M. Research* **8**:309, 1902.

Adrianoff, N., and Ansbacher, S.: *Deutsche med. Wochenschr.* **56**:357, 1930.

Albot, G.: *Ann. d'anat. path.* **8**:435, 1931.

- Anderson, R. M.: Arch. Path. **14**:335, 1932.  
Assmann, H.: München. med. Wehnschr. **78**:221, 1931.  
Baginsky, A.: Arch. f. Kinderh. **65**:324, 1916.  
Balan, N. P.: Beitr. z. path. Anat. u. z. allg. Path. **76**:198, 1927.  
Barbour, H. G., and Fisk, M. E.: J. Pharmacol. & Exper. Therap. **48**:341, 1933.  
Beattie, J. M., and Dickson, W. E. C.: Text-Book of Pathology, St. Louis, C. V. Mosby Company, 1926, p. 803.  
Beaver, D. C., and Pemberton, J. de J.: Ann. Int. Med. **7**:687, 1933.  
— and Robertson, H. E.: Am. J. Path. **7**:237, 1931.  
Bischoff, M.: Ztschr. f. exper. Path. u. Therap. **11**:445, 1912.  
Bolliger, A., and Inglis, K.: J. Path. & Bact. **36**:19, 1933.  
Böllman, J. L., and Mann, F. C.: Ann. Int. Med. **5**:699, 1931.  
Bortin, A.: M. J. & Rec. **132**:228, 1930.  
Boyd, William: Text-Book of Pathology, Philadelphia, Lea & Febiger, 1932, p. 489.  
Chalatow, S. S.: Beitr. z. path. Anat. u. z. allg. Path. **57**:85, 1914.  
Choisser, R. M., and Wilson, P.: U. S. Nav. M. Bull. **26**:354, 1928.  
Churchill, T. P., and Van Wagoner, F. H.: Proc. Soc. Exper. Biol. & Med. **28**: 581, 1931.  
Davidson, J.: J. Path. & Bact. **26**:127, 1923; **28**:621, 1925.  
Davis, N. C.: J. M. Research **44**:601, 1923-1924.  
— and Whipple, G. H.: Arch. Int. Med. **23**:711, 1919; **27**:679, 1921.  
Domagk, G.: Ztschr. f. Krebsforsch. **29**:302, 1929.  
Ely, T.: Boston M. & S. J. **170**:542, 1914.  
Fahr: Verhandl. d. deutsch. path. Gesellsch. **12**:162, 1909; Virchows Arch. f. path. Anat. **205**:397, 1911.  
Fiessinger, N.: Compt. rend. prem. conf. internat. de path. géog., 1922, p. 627.  
— and Wolf, M.: Compt. rend. Soc. de biol. **87**:627, 1922.  
— Wolf, M., and Blum, G.: ibid. **87**:19, 1922.  
Findlay, G. M.: Brit. J. Exper. Path. **5**:92, 1924.  
Fischer, B.: Zentralbl. f. Path. **33**:231, 1922.  
Fischler, F.: Deutsches Arch. f. klin. Med. **93**:427, 1908.  
Fishback, F. C.: Arch. Path. **7**:955, 1929.  
Flinn, F. B., and Von Glahn, W. C.: J. Exper. Med. **49**:5, 1929.  
Friedenwald, J.: J. A. M. A. **45**:780, 1905.  
Gardner, George H., et al.: Bull. Johns Hopkins Hosp. **36**:107, 1925.  
Ghon, A.: Leberzirrhose, in Aschoff, L.: Pathologische Anatomie, ed. 7, Jena, Gustav Fischer, 1928, vol. 2, p. 886.  
Goodpasture, E. W.: J. A. M. A. **76**:1545, 1921.  
Grote: Inaug. Dissert., Berlin, 1912.  
Grover, A. L.: J. A. M. A. **61**:458, 1913; Arch. Int. Med. **17**:193, 1916.  
Gye, W. E., and Purdy, W.: Brit. J. Exper. Path. **5**:238, 1924.  
Haban, G.: Beitr. z. path. Anat. u. z. allg. Path. **92**:88, 1933.  
Hall, E. M., and Butt, E. M.: Arch. Path. **6**:1, 1928.  
— and MacKay, E. M.: Am. J. Path. **7**:327 and 343, 1931.  
Handovsky, H.; Schultz, H., and Staemmler, M.: Arch. f. exper. Path. u. Pharmakol. **110**:265, 1925-1926.  
Heitzmann, O.: Arch. f. Dermat. u. Syph. **152**:344, 1926.  
Henoch, E.: Charité-Ann. **13**:636, 1887.  
Herkel, W.: Beitr. z. path. Anat. u. z. allg. Path. **85**:513, 1930.  
Higgins, G. M., and Anderson, R. M.: Arch. Path. **12**:186, 1931; **14**:42, 1932.

- Higgins, G. M.; Mann, F. C., and Priestley, J. T.: *ibid.* **14**:491, 1932.  
 —and Priestley, J. T.: *ibid.* **13**:573, 1932.
- Huguenin, R.; Neumours-Auguste, and Albot, G.: *Ann. d'anat. path.* **9**:263, 1932.
- Hurst, E. W., and Hurst, P. E.: *J. Path. & Bact.* **31**:303, 1928.
- Isobe, K.: *Mitt. a. d. Grenzgeb. d. Med. u. Chir.* **27**:750, 1914.
- Jaffé, R.: *Frankfurt. Ztschr. f. Path.* **24**:241, 1920; *Centralbl. f. allg. Path. u. path. Anat.* **31**:57, 1920.
- de Josselin de Jong: *Compt. rend. prem. conf. internat. de path. géog.*, 1931, p. 38.
- Karsner, H. T.: *Human Pathology*, Philadelphia, J. B. Lippincott Company, 1929, p. 698.
- Kelly, A. O. J.: *Am. J. M. Sc.* **130**:951, 1905.
- Kretz, R.: *Wien. klin. Wchnschr.* **7**:365, 1894; **13**:271, 1900; *Ergebn. d. allg. Path. u. path. Anat.* **8**:473, 1904; *Verhandl. d. deutsch. path. Gesellsch.* **8**:54, 1904; *Internat. Clin.* **3**:289, 1905.
- Kryle, J., and Schopper, K. J.: *Virchows Arch. f. path. Anat.* **215**:309, 1914.
- Kuczynski, M. H.: *Beitr. z. path. Anat. u. z. allg. Path.* **65**:315, 1919.
- Lacquet, A. M.: *Arch. Path.* **14**:164, 1932.
- Lamson, P. D., and Wing, R.: *J. Pharmacol. & Exper. Therap.* **29**:191, 1926.
- Leitman, G.: *Virchows Arch. f. path. Anat.* **261**:767, 1926.
- Lissauer, M.: *Deutsche med. Wchnschr.* **39**:18, 1913; *Berl. klin. Wchnschr.* **51**: 114 and 159, 1914; *Virchows Arch. f. path. Anat.* **217**:56, 1914.
- Longcope, W. T.: *Tr. A. Am. Physicians* **28**:497, 1913.
- Love, J. G.: *Arch. Path.* **14**:637, 1932.
- MacCallum, W. G.: *Text-Book of Pathology*, ed. 5, Philadelphia, W. B. Saunders Company, 1932, p. 310.
- McDonald, S. Jr.: *Brit. J. Ven. Dis.* **8**:263, 1932.
- McIndoe, A. H.: *Arch. Path.* **5**:23, 1928.
- MacMahon, H. E.: *Am. J. Path.* **7**:77, 1931.  
 —and Mallory, F. B.: *ibid.* **7**:299, 1931.
- McNee, J. W.: *Croonian Lecture*, *Brit. M. J.* **1**:1111, 1932.
- MacNider, W. de B.: *J. Pharmacol. & Exper. Therap.* **49**:100, 1933.
- Macchiarulo, O.: *Frankfurt. Ztschr. f. Path.* **34**:37, 1926.
- Mallory, F. B.: *Am. J. Path.* **1**:117, 1925; **7**:570, 1931; **9**:557, 1933; *Arch. Int. Med.* **37**:336, 1926.  
 —and Parker, F.: *Am. J. Path.* **7**:351, 1931.  
 —Parker, F., and Nye, R. N.: *J. M. Research* **42**:461, 1921.
- Mann, F. C., and Bollman, J. L.: *Arch. Path.* **1**:681, 1926; *Compt. rend. prem. conf. internat. de path. géog.*, 1931, p. 190.  
 —and Graham, A. S.: *Internat. Abstr. Surg.*, Sept. 1928, p. 176.
- Fishback, F. C.; Gay, J. G., and Green, G. F.: *Arch. Path.* **12**:787, 1931.
- Marine, D., and Lenhart, C. H.: *Arch. Int. Med.* **8**:265, 1911.
- Martin, J. F.: *Ann. de méd.* **21**:89, 1927.
- Meyer, E., and Heubner, W.: *Biochem. Ztschr.* **206**:212, 1929.
- Midorikawa, B.: *Tr. Jap. Path. Soc.* **15**:98, 1925.
- Moon, V. H.: *Am. J. M. Sc.* **177**:681, 1929; *Arch. Path.* **13**:691, 1932; *Geographic Variations in the Occurrence of Cirrhosis*, *Compt. rend. second. conf. internat. de path. géog.*, to be published.
- Muir, R.: *Text-Book of Pathology*, London, Edward Arnold & Co., 1929, p. 546.
- Murayama, K.: *Tr. Jap. Path. Soc.* **11**:41, 1921; **13**:81, 1923.
- Ogata, S.: *J. M. Research* **40**:103, 1919-1920.
- O'Leary, P. A.; Snell, A. M., and Bannick, E. G.: *J. A. M. A.* **90**:1856, 1928.

- Ophüls, W.: Proc. Soc. Exper. Biol. & Med. **8**:75, 1910.
- Opie, E. L.: J. M. Research **12**:147, 1904; J. Exper. Med. **12**:367, 1910.
- and Alvord, L. B.: J. A. M. A. **62**:895, 1914; **63**:136, 1914.
- Oshima, F., and Siebert, P.: Beitr. z. path. Anat. u. z. allg. Path. **84**:106, 1930.
- Öttenberg, R., and Abramson, H. A.: J. A. M. A. **84**:800, 1925.
- Pearce, R. M.: J. M. Research **12**:329, 1904; **15**:99, 1906; J. Exper. Med. **8**:64, 1906.
- Permar, H. H., and Goering, H. D.: Arch. Int. Med. **52**:398, 1933.
- Polson, C. J.: Brit. J. Exper. Path. **10**:241, 1929; **14**:73, 1933; J. Path. & Bact. **34**:5, 1931.
- Raab, W., and Terplan, C.: Med. Klin. **19**:1154, 1923.
- Rao, P. K.: Beitr. z. path. Anat. u. z. allg. Path. **87**:599, 1931.
- Reichle, H. S.: Arch. Int. Med. **49**:215, 1932.
- Rolleston, H. D.: Oxford System of Medicine, New York, Oxford University Press, 1927, vol. 3, p. 315.
- and McNee, J. W.: Diseases of the Liver, Gall-Bladder and Bile-Ducts, New York, The Macmillan Company, 1929, p. 215.
- Rosenthal, S. M.: J. Pharmacol. & Exper. Therap. **38**:291, 1930.
- Ross, J. B.: Canad. M. A. J. **24**:632, 1931.
- Rössle, R.: Virchows Arch. f. path. Anat. **291**:1, 1933.
- Saltykow, S.: Verhandl. d. deutsch. path. Gesellsch. **14**:228, 1910.
- Schafir, M.: Virchows Arch. f. path. Anat. **213**:41, 1913.
- Schindel, L.: Beitr. z. path. Anat. u. z. allg. Path. **87**:768, 1913.
- Schirokogeroff, I. I.: First Allruss, Path. Congr. Petrograd, 1923; Ref. Centralbl. f. allg. Path. u. path. Anat. **35**:74, 1924.
- Schultz, E. W.; Hall, E. M., and Baker, H. V.: J. M. Research **44**:207, 1923.
- Scott, E., and Helz, M. K.: Am. J. Hyg. **16**:865, 1932.
- Sherwood, K. K., and Sherwood, H. H.: Arch. Int. Med. **48**:82, 1931.
- Silbergleit, H., and Föckler: Ztschr. f. klin. Med. **88**:333, 1919.
- Smetana, Hans: Arch. Path. **15**:175, 330 and 516, 1933.
- Sollmann, T. H.: Manual of Pharmacology, Philadelphia, W. B. Saunders Company, 1926.
- Stephenson, G. W.: Arch. Path. **14**:484, 1932.
- Testoni, P.: Clin. med. ital. **57**:428, 1926.
- Van der Schueren, G.: Compt. rend. Soc. de biol. **109**:982, 1932.
- Van Wagoner, F. H., and Churchill, T. P.: Arch. Path. **14**:860, 1932.
- Villaret, M.; Bertraud, I.; Justin-Besançon, L., and Even, R.: Ann. de méd. **31**:334, 1932.
- Wallace, R. P.: Proc. Soc. Exper. Biol. & Med. **24**:598, 1927.
- Weller, C. V.: Tr. A. Am. Physicians **45**:71, 1930.
- Whipple, G. H., and Sherry, J. A.: Bull. Johns Hopkins Hosp. **20**:278, 1909.
- Williamson, C. S., and Mann, F. C.: Am. J. Physiol. **65**:267, 1923.
- Yenikomishian, H. A.: Nonalcoholic Cirrhosis of the Liver in Lebanon and Syria, J. A. M. A. **103**:660 (Sept. 1) 1934.

## Notes and News

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**Inspection of Medical Schools.**—H. G. Weiskotten, dean and professor of pathology in the College of Medicine, Syracuse University, has been granted leave of absence for two years to carry on inspection of medical colleges in the United States under the Council of Medical Education and Hospitals of the American Medical Association, the Association of American Medical Colleges and the Federation of State Medical Boards. The work of inspection will begin with the opening of the college sessions for 1934-1935.

**Harvard Statement About Patents.**—According to *Science*, the Harvard Corporation has issued the following statement: "In the opinion of the Faculties of Medicine, Public Health, Engineering and Arts and Sciences, the patenting by members of the university of discoveries or inventions bearing on matters of health and therapeutics is undesirable. The president and fellows of Harvard College, therefore, have adopted the rule that no patents primarily concerned with therapeutics or public health may be taken out by any member of the university, except with the consent of the president and fellows, nor will such patents be taken out by the university itself unless they be dedicated to the public. The president and fellows will provide legal advice to anyone who desires steps to be taken to prevent the patenting by others of his discoveries or inventions."

**Cancer Research at University in Jerusalem.**—Some \$200,000 has been donated for cancer research at the Hebrew University in Jerusalem. Ludwig Halberstaedter, formerly of the Cancer Institute in Berlin, has been nominated head of the department of radiobiology; H. A. Krebs, formerly of Berlin and now at Cambridge University, England, has been nominated as head of the department of physiologic chemistry, and Leonid Doljansky, formerly of the Institute of Pathology of the University of Berlin and now at the University of Copenhagen, has been nominated as head of the department for the study of cells and tissues and morbid anatomy.

**Institute of Legal Medicine in Lille.**—A new institute of legal medicine has been opened at Lille, France. Facilities are provided for ordinary medicolegal work as well as for criminal anthropology, penology, occupational diseases and study of delinquents.

# Abstracts from Current Literature

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## Pathologic Anatomy

THE ARTERIAL BLOOD VASCULAR DISTRIBUTION TO THE LEFT AND RIGHT VENTRICLES OF THE HUMAN HEART. LOUIS GROSS and M. A. KUGEL, Am. Heart J. 9:165, 1933.

By means of a new technic the injection of a standardized barium sulphate gelatin suspension through the coronary arteries has been made possible. Immediately after injection the hearts are fixed in 10 per cent formaldehyde saline solution. After fixation the hearts are cut into transverse slices, 7 mm. thick, and then roentgenograms are made whereby it is possible to study the distribution of the vessels in the myocardium and to compare unit areas in the left and right ventricles. It has been definitely demonstrated that early in life there is a predominance of right ventricular vessels, that there soon follows an equalization of the vascular supply, which lasts for approximately one decade, and that thereafter the vascular bed in the myocardium of the left ventricle assumes an increasing preponderance over that of the right. It has also been shown that the septal anastomoses increase very conspicuously in their extent, particularly after the third decade in life. Both of these observations confirm the earlier report by Gross in this field.

### AUTHORS' SUMMARY.

CORONARY OCCLUSION DUE TO METASTASES FROM CARCINOMA OF THE BREAST. T. A. PEPPARD and L. M. LARSON, Am. Heart J. 9:265, 1933.

Undoubtedly in this case death was caused by an unusual sequence of events which terminated in occlusion of the coronary arteries. Late metastases from an adenocarcinoma of the breast, which had been removed by radical resection five years previously, appeared in a very unexpected location, namely, in the epicardium surrounding the coronary arteries. The mechanism of the localization of the metastases in this particular place is obscure, although unquestionably the blood stream was the medium through which the transfer of malignant cells took place. While in this person a certain amount of arteriosclerotic thickening could be expected because of her age, it probably would have caused few symptoms. However, with the added insult of gradual obstruction of the arterial supply of blood to the heart by the carcinomatous tissue, symptoms and signs of cardiac failure became more and more prominent until death supervened. Metastatic tumors of the heart of various types, while they are comparatively rare, have been reported from time to time, but none has been found which corresponds to the type of lesion described in this case.

### AUTHORS' SUMMARY.

XANTHOMATOSIS. KATHARINE K. MERRITT and BERYL H. PAIGE, Am. J. Dis. Child. 46:1368, 1933.

A case of generalized xanthomatosis (Schüller-Christian's syndrome) with characteristic skeletal changes, exophthalmos, diabetes insipidus and cutaneous lesions is reported. The most unusual feature was the severe cutaneous involvement. Pathologic changes characteristic of the disease were found in the bones, dura mater, hypophysis, infundibulum, periosteum, orbits, skin, thymus, pleura, lungs, heart, peritoneum, spleen, liver, pancreas, lymph glands and retroperitoneal adipose tissue. In the spleen and lymph glands there was extensive hyperplasia of the reticulo-endothelial cells, without evidence of lipoid storage, but in many sites, such as the dura, bones, lungs and thymus, there were typical xanthomatous lesions.

In the skin the corium contained localized accumulations of large mononuclear cells similar morphologically to the reticulo-endothelial cells found in other regions, but in the cutaneous infiltrations few cells contained fat or lipoids.

#### AUTHORS' SUMMARY.

#### INTESTINAL LESIONS ASSOCIATED WITH AMEBIC AND BALANTIDIAL INFECTION.

H. L. RATCLIFFE, Am. J. Hyg. 19:68, 1934.

Pathogenic species of Endamoeba and Balantidium have identical effects on the mucosa of the large intestine in susceptible hosts. The primary and perhaps the total effect of the organisms is merely necrosis. They enter the tissue only after necrosis has taken place, except as a postmortem or late antemortem change. The protozoa apparently elaborate a cytotoxic or necrotizing substance that causes tissue degeneration. Lesions are invaded by intestinal bacteria also. Hence extension of tissue necrosis may be due to a combination of factors, and the inflammatory response may not be attributed to the parasites alone. Resistance of the host must be impaired before the intestinal tissues are subject to necrosis. In either balantidiasis or amebiasis, lesions of the intestinal wall originate in the mucous surface. Surface and deep ulcers are genetically the same, except when the latter result from bacterial abscesses of the follicles. The lymphoid tissues of the intestinal wall, because of their anatomic position and make up, are important foci for extension of the disease to the submucosa. Postmortem penetration of tissue by Balantidium and Endamoeba accounts for the fact that isolated organisms and groups of organisms are found deep in the intestinal wall. Balantidium also can invade cut edges of the large intestine at autopsy.

#### AUTHOR'S SUMMARY AND CONCLUSIONS.

#### RHEUMATIC PERITONITIS. L. J. RHEA, Am. J. Path. 9:719, 1933.

Except for the fact that the peritoneal lesions are diffuse rather than focal and sharply outlined, they are similar in every way to the so-called Aschoff bodies found in the heart in this case and in other cases of rheumatic fever. I believe that these lesions are specific, resulting from the action on the peritoneum of the causative agent of the disease. It seems probable that the involvement of the peritoneum in rheumatic fever is more common than is generally thought. If careful microscopic examination is made of tissues from various regions of the peritoneum in instances of rheumatic fever, especially in the severe cases, the specific lesion may be found to be fairly common.

#### AUTHOR'S CONCLUSIONS.

#### NODULAR LESIONS OF THE PERITONEUM. S. R. HAYTHORN, Am. J. Path. 9:725, 1933.

Nearly thirty cases of nodular lesions of the peritoneum which were of interest from the standpoint of surgical diagnosis have been collected, and an outline has been prepared to facilitate the more rapid classification of such lesions.

#### AUTHOR'S SUMMARY.

#### THE BONE MARROW IN SPRUE ANEMIA. C. P. RHOADS and W. B. CASTLE, Am. J. Path. 9:813, 1933.

Observations on the bone marrow in sprue anemia, made on tissue obtained at biopsy in different stages of the disease, show that the changes are similar to those of pernicious anemia. During relapse the essential change is a proliferation of megaloblasts and a suppression of maturation to the normoblast stage. During remission the marrow tends to return to normal with a great increase in the number of normoblasts and mature red cells in the marrow. Phagocytosed erythrocytes were observed in the bone marrow removed at autopsy, but not in that removed during life.

#### AUTHORS' CONCLUSIONS.

CHANGES IN THE BRAIN DUE TO ARSPHENAMINE. J. H. GLOBUS and S. W. GINSBURG, Arch. Neurol. & Psychiat. 30:1226, 1933.

A case of syphilis of the brain and one of so-called acute multiple sclerosis were studied. In the former ringlike hemorrhages were present in the optic thalamus, cornu ammonis, tegmentum of the midbrain, pons and cerebellar cortex. In the second case the hemorrhages were scattered in the hemispheres and were especially prominent in the temporal lobes, internal capsule, midbrain and pons; a few were seen in the medulla and spinal cord. In the opinion of the authors, the hemorrhages were capillary and were due to damage of the vascular endothelium.

GEORGE B. HASSIN.

MYOTONIC DYSTROPHY. M. KESCHNER and C. DAVISON, Arch. Neurol. & Psychiat. 30:1259, 1933.

The histologic changes in a white man, aged 40, in whom the disease lasted eleven years, were: increase in connective tissue in the anterior lobe of the pituitary gland with degeneration of the eosinophilic cells; hyalinization of the interstitial tissue of the thyroid gland; central fibrosis of the parathyroid glands; fibrosis of the fascicular layer of the suprarenal glands, and atrophy of the seminiferous tubules of the testes with increase in Leydig's cells. The muscles, smooth and striated, had an increase in the sarcolemmic nuclei with loss of striations and hyalinization in many instances. In the brain, a spongioblastoma of the corpus callosum, the size of a pea, was found which had given no clinical symptoms. The ganglion cells of the hypothalamic and paraventricular nuclei and of the lateral columns of the spinal cord showed marked chromatolysis, tumefaction and disintegration, while the thalamus and the nuclei of the medulla presented milder changes. In the sister of this patient who also suffered from myotonic dystrophy a biopsy revealed a proliferation of connective tissue in the muscles, which were replaced by fat and chains of sarcolemmic nuclei. The primary changes are not in the sympathetic nervous system, but in the muscles; the changes in the lateral horn cells are secondary.

GEORGE B. HASSIN.

PATHOGENESIS OF APOPLECTIC CEREBRAL HEMORRHAGE. T. VON LEHOCZKY, Beitr. z. path. Anat. u. z. allg. Path. 92:132, 1933.

To determine whether apoplectic cerebral hemorrhage is the result primarily of vascular disease, as is generally believed, or of disease of the cerebral parenchyma, as some recent observers maintain, von Lehoczky examined histologically the brains of fourteen persons who died of such hemorrhage. When death occurred within a few hours after the hemorrhage, there were observed fibrillar separation and marked hypertrophy of the glia in and about the area of hemorrhage. He believes that this parenchymatous lesion was present before the hemorrhage occurred and was a factor in it. Vascular disease was present in all the brains. He concludes that cerebral hemorrhage may be due to disease of the parenchyma, to vascular disease or to arterial hypertension, the three factors often operating together.

O. T. SCHULTZ.

ABLATION OF THE PLEURA AND RUPTURE OF PULMONARY TISSUE. F. ORSÓS, Beitr. z. path. Anat. u. z. allg. Path. 92:173, 1933.

Orsós reports nine examples of ablation of the pleura or rupture of pulmonary tissue. The cases represent a variety of clinical conditions, but in all there was round cell infiltration of the lung or pleura. The tearing of the inflamed and weakened tissue was a terminal event and was the result of dyspnea or violent respiratory efforts. The air-filled spaces formed beneath the pleura or in the lung are not to be confused with those seen in emphysema.

O. T. SCHULTZ.

PECULIAR MALFORMATION OF THE PULMONARY VEINS. H. REBENSBURG, Frankfurt. Ztschr. f. Path. 44:137, 1932.

A male infant died suddenly about three months after birth. The left ventricle was much smaller than the right. The aorta showed no changes, and the ductus arteriosus was obliterated. There was no communication between the left auricle and the lungs, the pulmonary veins not being in their normal location. The foramen ovale was wide open and round, measuring 5 mm. in diameter, and the right auricle was three times as large as the left. Into the right auricle opened the coronary veins and the superior and inferior venae cavae, in addition to another vein which extended from the right upper lobe of the lung into the auricle. Another pulmonary vein took its course from the lower lobe of the right lung to the lower lobe of the left lung, forming a bridge between these lobes. This vein also extended throughout the entire left lung to the region of the hilus, collecting smaller tributaries, and opened into the superior vena cava just above the right auricle. The pulmonary arteries were normal. The arterial blood took the following courses from the lung to the heart; from the right upper lobe into the right auricle; from the right lower lobe to the left lung and from there to the superior vena cava and also into the right auricle; from the right auricle through the patent foramen ovale into the left auricle.

O. SAPHIR.

SO-CALLED SEPTIC INFLAMMATION OF THE SPLEEN. K. VON WOLFF, Frankfurt. Ztschr. f. Path. 44:161, 1932.

By injecting foreign proteins (milk), Wolff attempted to produce changes in the spleen similar to those seen in cases of sepsis. The conclusion is reached that so-called septic inflammation of the spleen is not the direct result of the presence of micro-organisms but is caused by protein material set free by destruction of cells. Therefore, the terms toxic splenitis, septic splenitis, and the like are incorrect; "splenitis albuminotoxica" seems more significant.

O. SAPHIR.

TWO CASES OF AGRANULOCYTOSIS IN CHILDREN. R. GANSER, Frankfurt. Ztschr. f. Path. 44:329, 1932.

The important manifestations in these cases were hemorrhagic diathesis, enlargement of the liver and mastoiditis. Angina was present in one case. In both instances the blood showed marked reduction in the number of the red corpuscles and marked leukopenia, with relative lymphocytosis and thrombopenia. In addition, the second case revealed polychromatophilia of the red corpuscles and a moderate number of normoblasts. Several blood transfusions were given, resulting in an improvement of the blood picture. Gradually, however, the anemia and leukopenia recurred. Autopsy revealed hemorrhages into the skin, the serous and mucous membranes, the myocardium, the surface of the liver and kidneys and the subperiosteal space. In the first instance the lymph nodes in the region of the neck and hilus of the liver were moderately enlarged, while in the second case all the lymph nodes were enlarged. The mucosa covering the tonsils showed areas of hemorrhage and necrosis. The bone marrow of the femur was dark red in both instances. Histologic examination of the bone marrow revealed an almost complete disappearance of granulocytes and megakaryocytes. Erythrocytes, erythroblasts and a large number of cells that showed only a small amount of cytoplasm were present. The nuclei of the latter varied in size. These cells resembled microlymphocytes and mesolymphocytes. Also, cells apparently identical with the hemocytoblast (stem cell) of Maximow were recognized. The lymph nodes revealed microlymphocytes, mesolymphocytes and a few macrolymphocytes and hemocytoblasts. Occasionally eosinophilic myelocytes were also seen. The spleen contained microlymphocytes and mesolymphocytes, a few plasma cells, myelocytes and many partially destroyed erythrocytes. Small and large lymphocytes were present in the liver in addition to eosinophilic myelocytes, which were

abundant in the second case. In spite of the presence of many microlymphocytes and mesolymphocytes, Ganser does not believe that these two cases are characteristic of chronic (aleukemic) lymphadenosis. He believes that these cells are closely related to the hemocytoblasts and very likely are myeloid cells. It is possible that the stem cells and their derivatives are increased in number because of a pathologic irritation. Ganser concludes that severe damage, possibly combined with a constitutional factor, produces marked destruction and necrosis of the bone marrow and may be responsible for the picture of so-called panmyelophthisis. A less severe irritation may produce atrophy of the blood-forming organs. Another type of irritation, still less severe, may cause, in addition to atrophy, regenerative and compensatory blood formations in the bone marrow and other organs. He believes that the latter explanation holds for the two cases reported.

O. SAPHIR.

HISTOLOGIC CHANGES OF THE LYMPH NODES IN CASES OF LYMPHOGRANULOMATOSIS (HODGKIN'S DISEASE). H. HAMDI and SAIM-ALI, Frankfurt. Ztschr. f. Path. 44:338, 1932.

The lymph nodes of six persons showing Hodgkin's disease were examined. The small nodes revealed a marked increase in the number of lymphocytes and occasionally in the numbers of eosinophilic and plasma cells. These changes are thought to be the initial stages of Hodgkin's disease. In the enlarged lymph nodes were found a number of epithelioid cells, varying numbers of lymphocytes, polymorphonuclear neutrophilic and eosinophilic cells, plasma cells and giant cells, areas of hyalinization and connective tissue fibers. In addition, foci of necrosis were seen. The epithelioid cells were round, oval or sometimes polygonal. Their cytoplasm was abundant and occasionally revealed pseudopodia-like elongations. The nuclei were large, round and often lobulated. They were distinctly vesicular, revealed a fine chromatin network and granules, and contained one or two metachromatic nucleoli. Mitotic figures were present. Some of these cells were unusually large and revealed several nuclei. Cells of the latter type probably originated from the perithelial cells of blood vessels. It is also possible that they arose from the reticulo-endothelial system. It is likely that the giant cells resulted from the confluence of several epithelioid cells. In the larger and older nodes the strands of connective tissue extended from the capsule into the granulation tissue. The various stains for the presence of bacteria did not reveal uniform results. Occasionally, organisms of the Corynebacterium group could be demonstrated by the use of Gram's method. Fresh portions of lymph nodes transplanted into rabbits and mice grew in only one instance. The transplant contained bacteria and connective tissue fibers but showed no granulomatous changes.

O. SAPHIR.

FREQUENCY AND ORIGIN OF SO-CALLED ROUND CELL FOCI IN THE SUPRARENAL GLANDS. A. COSTA, Frankfurt. Ztschr. f. Path. 44:343, 1932.

Foci of round cells are found in the human suprarenal glands at every age. In children similar cells are isolated and in very small groups; they are more common in the zona reticularis. Later they extend into the zona fasciculata.

O. SAPHIR.

INCOMPLETELY FORMED CORPUS CALLOSUM REPLACED BY FATTY TISSUE. B. G. RUBINSTEIN, Frankfurt. Ztschr. f. Path. 44:379, 1932.

A series of anomalies were found in the brain of a 30 year old man who had had epileptic attacks since childhood. The corpus callosum was partly absent. There was hypoplasia of the middle portion of the fornix, of the choroid plexus and of the third ventricle. The septum pellucidum and the lyra were absent, and the columnae fornicis revealed an abnormal course. In place of the corpus

callosum fatty tissue was found, which is explained as a secondary process. Since fat was also present in the choroid plexus, the fat probably extended from there into the region of the corpus callosum.

O. SAPHIR.

**DELAYED THORIUM DIOXIDE NECROSIS OF LYMPH NODES.** T. NAEGELI and A. LAUCHE, Klin. Wchnschr. **12**:1730, 1933.

During three years' observation of a dog given intravenous injections of thorium dioxide there was no appreciable excretion of this compound. The dioxide was transported from the site of initial deposition into the regional lymph nodes, where it produced extensive necrosis. The further fate of the thorium dioxide is speculative. Probably some became encapsulated and some was carried into other lymph nodes.

AUTHOR'S SUMMARY.

**PENETRATION OF A GASTRIC ULCER INTO THE WALL OF THE HEART.** W. EHRHARDT, Virchows Arch. f. path. Anat. **289**:327, 1933.

A woman, aged 63, who had been ill for two months with symptoms referable to an inoperable carcinoma of the sigmoid flexure died suddenly of heart failure. Necropsy revealed, in addition to the carcinoma, an ulcer of the lesser curvature of the stomach that had penetrated the diaphragm and eroded the outer portion of the wall of the left ventricle. The heart was adherent to the pericardium only in this region. The ventricular wall was not perforated. Seven previously reported cases of this unusual complication of gastric ulcer are discussed.

O. T. SCHULTZ.

**SUBAORTIC DEFECT OF THE INTERVENTRICULAR SEPTUM OF THE HEART.** H. TESSERAUX, Virchows Arch. f. path. Anat. **289**:412, 1933.

The heart described was that of a boy, aged 12 years, who died of acute leukemia. The heart was enlarged to about three times its normal size. In the anterior upper part of the interventricular septum was an opening that measured 11 by 17 mm. on the side of the left ventricle, where it was situated below the right and left segments of the aortic valve. On the right side the opening was smaller and was at a considerable distance from the ostium of the pulmonic artery. The defect was situated above the conduction system. The few previously reported instances of a defect in this unusual situation are reviewed by the author. He ascribes the defect to an abnormally large embryonic interventricular foramen.

O. T. SCHULTZ.

**SOME UNUSUAL MALFORMATIONS.** F. GRÖGLER, Virchows Arch. f. path. Anat. **289**:430, 1933.

Four fetuses with craniorachischisis, two acardiac monstrosities and one phocomelus with the defect limited to one upper extremity are described. Craniorachischisis is ascribed by Grögler to the action of external factors. The phocomelus was a woman who died at the age of 21 of pulmonary tuberculosis. The forearm, two phalanges and part of the carpus were absent.

O. T. SCHULTZ.

### Microbiology and Parasitology

**PRENATAL TRICHINOSIS.** D. L. AUGUSTINE, Am. J. Hyg. **19**:115, 1934.

Observations on rabbits, rats, swine and a child born of trichinous mothers confirm the conclusion of Stäubli (1909) that prenatal infection does not occur in trichinosis.

AUTHOR'S CONCLUSION.

THE SELF-DISINFECTING POWER OF THE SKIN. L. ARNOLD and A. BART, Am. J. Hyg. 19:217, 1934.

Living bacteria placed on the skin rapidly disappear. They cannot be demonstrated by cultural or tinctorial methods thirty or forty minutes after contact with the skin. Attempts have been made to determine the presence of antigenic substances placed on the skin. *Bacillus enteritidis* and fresh egg-white placed on cutaneous surfaces and allowed to dry in the air could not be demonstrated in washings collected from such exposed surfaces. These antigens could be demonstrated in cornified epithelial scrapings mechanically removed from such exposed skin. The human skin possesses a power of destroying exogenous bacteria placed in contact with it. Desiccation is not a major factor in this mechanism of controlling the viable bacterial flora on a clean cutaneous surface. Evidence is presented to indicate that keratin may play a rôle in the removal of bacterial as well as other antigens from human skin.

AUTHORS' SUMMARY.

THE CLINICAL SIGNIFICANCE OF *B. COLI HEMOLYTICUS*. WALTER L. NILES and JOHN C. TORREY, Am. J. M. Sc. 187:30, 1934.

Strains of the colon bacillus possessing hemolytic properties are frequently isolated from human feces (31 to 45 per cent of stools). They are somewhat more often found and in larger numbers in feces from patients with disorders relating to the digestive tract. There is no distinctive difference as regards morphology or cultural characteristics in the hemolytic strains of the colon bacillus isolated from the stools of normal and sick people, although those from the former group have seldom exhibited any virulence. We have been successful in relieving the symptoms of toxemia by autogenous vaccines. *Bacillus coli-haemolyticus* has thus been eliminated from the feces in 93 per cent of our cases. It has reappeared in 3 per cent after one year or longer. Vaccination is the only method for eliminating *B. coli-haemolyticus* from the human feces which we have found effective.

AUTHORS' SUMMARY.

CONJUGAL SYPHILIS—A STATISTICAL STUDY. HENRY B. DECKER, Am. J. M. Sc. 187:111, 1934.

There is presented an investigation of the records of 376 families in which syphilis was present in at least one spouse; 60 husbands and 98 wives were syphilitic with nonsyphilitic spouses. In 218 families both husband and wife were syphilitic. It is concluded that the type of syphilis depends on the individual rather than on any special strain of spirochetes. Late syphilis is relatively non-infectious. Infectivity varies inversely with the duration of the disease.

AUTHOR'S SUMMARY.

THE ETIOLOGY OF MUMPS. C. D. JOHNSON and E. W. GOODPASTURE, J. Exper. Med. 59:1, 1934.

From four of six specimens of saliva taken in six cases of mumps in the early stages of the disease a filtrable cytotropic virus has been obtained which induces in rhesus monkeys, following its injection into the parotid gland through Stensen's duct, an acute nonsuppurative parotitis analogous to mumps. This virus has not been found in normal saliva, nor does it correspond to any known virus with which we are familiar. The virus is free from demonstrable micro-organisms including spirochetes. It is judged that this virus is the causative agent in mumps.

AUTHORS' CONCLUSIONS.

THE SIGNIFICANCE OF HORMONAL FACTORS IN EXPERIMENTAL POLIOMYELITIS.  
C. W. JUNGEBLUT and E. T. ENGLE, *J. Exper. Med.* **59**:43, 1934.

Adult rhesus monkeys possess frequently a greater resistance to experimental infection with the virus of poliomyelitis than young monkeys, as indicated by the prolongation of the incubation period and by the content of feeble neutralizing substances in the normal serum. Virucidal substances can be demonstrated in the normal serum of subadult chimpanzees. Immature monkeys prepared with anterior lobe pituitary hormones and the like principles from the urine of pregnant women are only in exceptional instances protected against intracerebral infection with poliomyelic virus. However, the serum of such prepared animals frequently acquires the property of inactivating the virus in vitro. Active immunization with the live virus renders the immunized monkeys resistant to intracerebral infection in a higher percentage of the cases than does preparation with hormones. The relative merits of nonspecific enhancement of resistance and of specifically acquired immunity are discussed in their relation to the probable mechanism of resistance to poliomyelitis in man.

AUTHORS' CONCLUSIONS.

SIMILARITIES BETWEEN EXPERIMENTAL SCURVY COMBINED WITH INFECTION AND RHEUMATIC FEVER. J. F. RINEHART, C. L. CONNOR and S. R. METTIER, *J. Exper. Med.* **59**:97, 1934.

In the guinea-pig, chronic scurvy with superimposed infection (beta hemolytic streptococcus), and to a lesser extent chronic scurvy alone, produces an arthropathy with striking pathologic similarities to that of rheumatic fever and the closely allied condition of rheumatoid arthritis. Considerable significance is attached to the widespread occurrence in the animal subjected to scurvy and infection, and to a lesser extent in scurvy alone, of lesions similar to if not identical with the fibrinoid degeneration which has been considered the fundamental lesion of rheumatic fever. A subcutaneous nodule essentially similar to the subcutaneous nodules of rheumatic fever was observed in one experimental animal. Attention is called to a group of general pathologic changes frequently observed in rheumatic fever which were also found in the animals subjected to scurvy and infection. These include degenerative changes in skeletal muscle, focal necrosis in the liver, fibrosis of the malpighian bodies in the spleen, erythrophagocytosis in the lymph nodes and focal lymphocytic accumulations in the kidneys. The problem of hemorrhage is considered. It is suggested that a scorbutic state may be the basis of the hemorrhagic manifestations common to the acute phases of rheumatic fever. The unsatisfactory nature of previous attempts to reproduce rheumatic fever is noted. The lesions produced by subjecting the guinea-pig to the combined influence of scurvy and infection are considered to be fundamentally similar in character and distribution to those of rheumatic fever. The observations recorded are believed to offer evidence that the disease known as rheumatic fever may be the result of the combined influence of scurvy and infection, and that scurvy may constitute the rheumatic tendency in which the added factor of infection causes the development of rheumatic fever or possibly the closely allied condition of rheumatoid arthritis.

ACUTE ASCENDING MYELITIS FOLLOWING A MONKEY'S BITE. A. B. SABIN and A. M. WRIGHT, *J. Exper. Med.* **59**:115, 1934.

A case of acute ascending myelitis which followed the bite of an apparently normal rhesus monkey is described. The absence of perivascular demyelination removes the case from the realm of acute disseminated encephalomyelitis and establishes it more or less definitely as one of primary acute infectious myelitis. An extremely important pathologic feature was the focal necrosis in the spleen, suprarenal glands and regional lymph nodes. Attempts to transmit the disease to rhesus monkeys, dogs, mice and guinea-pigs, employing glycerinated organs from the patient, proved unsuccessful. By inoculations of rabbits the presence of a strongly neurotropic filtrable virus was demonstrated in the patient's brain, cord

and spleen. Following intracutaneous injection of the virus as derived either from brain and cord or from spleen, an experimental disease developed in rabbits which strikingly resembled the human disease in the character of the local lesion, the period of incubation, the development of urinary retention, the flaccid paralysis of the posterior extremities with cephalad progression, the death by respiratory failure, and finally the observation of focal necrosis in the spleen, suprarenal glands and liver. In attempting to establish the identity of this virus (the B virus), a consideration of its biologic properties excludes the viruses of poliomyelitis, rabies, vaccinia and virus III disease of rabbits and the other viruses which are known to produce similar intranuclear inclusions, except perhaps that of herpes. Although the relationship between the B virus and the virus of herpes must still be determined by cross-immunity tests the B virus has been shown to possess certain properties which warrant consideration of it as a distinct entity.

FROM THE AUTHORS' SUMMARY.

THE CONVERSION OF TYPHUS STRAINS. H. MOOSER and others, J. Exper. Med. 59:137, 1934.

It is concluded from our experiments that there does not exist any real difference between the virus of historic Old World typhus and the murine New World typhus. Both are considered to be of murine origin. The murine strains represent the original form of the virus of typhus, whereas the epidemic strains are the result of a prolonged propagation in the cycle man-louse-man.

FROM AUTHORS' SUMMARY AND CONCLUSIONS.

THE VIRUSES OF VESICULAR STOMATITIS AND EQUINE ENCEPHALOMYELITIS. P. K. OLITSKY and others, J. Exper. Med. 59:159, 1934.

The viruses are similar in many biologic properties. In view of the fact that the horse is the natural host for both it is suggested that they may be generically related. They are not, of course, identical, since cross-immunity does not exist. This, however, does not exclude the possibility of a generic relationship, for there are at least three immunologically distinct types of foot and mouth diseases, two of vesicular stomatitis, and two of equine encephalomyelitis.

FROM AUTHORS' SUMMARY.

THE SURVIVAL OF NEUROTROPIC YELLOW FEVER VIRUS IN TESTICULAR TISSUES. W. LLOYD and A. F. MAHAFFY, J. Immunol. 25:471, 1933.

Neurotropic yellow fever virus of the French strain and of mouse origin has been transferred through nine passages in the testicular tissues of albino mice. Virus isolated from the sixth and seventh passages in the testes of mice has been demonstrated to give a specific immunologic reaction as yellow fever virus in passive immunity tests. Testicular virus of mouse origin when injected subcutaneously into a rhesus monkey resulted in the immunization of the animal, with the development of only a retarded and mild febrile reaction.

AUTHORS' SUMMARY.

THE INFECTIVITY OF NEUROTROPIC YELLOW FEVER VIRUS FOR ANIMALS. G. M. FINDLAY, J. Path. & Bact. 38:1, 1934.

After intracerebral inoculation neurotropic yellow fever virus produces an encephalitis not only in mice and guinea-pigs but also in field voles and red squirrels. Cats, ferrets, rabbits, rats, golden hamsters, wood voles, bank voles, pigeons, hens and canaries are not susceptible and suffer neither encephalitic changes nor viscerotrophic lesions. Little or no multiplication occurs in the brain of a nonsusceptible species such as the rat. The neurotropic virus is capable of surviving for a few days in the brains of nonsusceptible species. Immune bodies for neuro-

tropic yellow fever virus are not present in the blood of nonsusceptible species before inoculation, but may subsequently appear in the blood.

#### AUTHOR'S CONCLUSIONS.

GENERALIZED VACCINIA IN MAN. J. H. DIBLE and H. H. GLEAVE, *J. Path. & Bact.* **38**:29, 1934.

A study has been made of the lesions in a case of generalized vaccinia in a child aged 3 months who died on the twenty-sixth day after vaccination. The skin pocks appeared clearly as primary epithelial necrosis without any granulomatous changes in the corium. The visceral lesions, like the epidermal, were purely necrotic without hemorrhage and with only a minimum reactive change. There were no vaccinal lesions found in the lungs, the brain or the suprarenal glands.

FUNGI FROM BLASTOMYCOSIS. A. CASTELLANI and I. JACONO, *J. Trop. Med.* **36**:297, 1933.

Blastomycosis may be caused by many different fungi. Cultures of the fungi described in the paper will be supplied to interested workers by the authors (Ross Institute and Hospital for Tropical Diseases, London).

EXPERIMENTAL SYPHILIS IN LABORATORY ANIMALS. J. VAN HAELST, *Arch. internat. de méd. expér.* **8**:543, 1933.

Syphilis ran a typical course in rabbits and guinea-pigs regardless of the quantities of spirochetes in the inoculated material. Evidences of syphilis became apparent more quickly if syphilitic lymph nodes were employed for inoculation than if syphilitic testis was used. In either case the period of incubation lengthened as the infective material was diluted. In the rabbit the potential infectiveness of syphilitic testis was greater than that of syphilitic inguinal or popliteal nodes, in the sense that testis was capable of provoking a primary lesion when used in a dilution ineffective in the case of the nodes. A striking difference in minimum infective dosages of these tissues could not be explained on the basis of the relative numbers of spirochetes contained. Syphilitic guinea-pig lymph node was peculiar in that it was much more infective than syphilitic rabbit testis. In the guinea-pig, more manifestly than in the rabbit, the period of incubation of the primary lesion was longer than usual if infected node, rather than testicle, was inoculated. Large doses of syphilitic material appeared to shorten the period of incubation, but minimum doses showed no tendency to lengthen it. Spirochetes were detected in 95.65 per cent of syphilitic rabbit nodes examined. Nodes draining active lesions were found to contain greater numbers of spirochetes than nodes from the region of healed lesions. Great numbers of spirochetes were found in the regional lymph nodes on the day following inoculation. In the clinically nonapparent syphilis of the mouse, spirochetes were seen not only in the superficial nodes but also in the spleen and brain. Spirochetes were most numerous in the lymphoid tissues of the mouse. In the rabbit, the minimum infective dose bore a direct relation to the quantities of spirochetes contained when tissues relatively poor in spirochetes were employed. The minimum infective dose of lymph node, a tissue poor in spirochete content, approached a constant value which was less than that required when other tissues containing many more spirochetes were employed. In infecting the rabbit with syphilis by inoculation of guinea-pig and mouse tissues, the factors of actual spirochete content and of specific infectivity of tissues poor in spirochetes seemed equally important in influencing the length of incubation of the primary lesions. Finally, it seemed that the presence of spirochetes was enough to explain the specific infectiveness of tissues which were low in actual spirochete content. Taken individually, spirochetes present in the primary lesion were probably less virulent than those in the regional nodes. The nodes apparently served to adapt the spirochete to live in the host, so that there was a rise in virulence.

**DETERMINATION OF TUBERCLE BACILLEMA IN CHILDREN. JOSEPH SIEGL,**  
**Beitr. z. Klin. d. Tuberk. 81:557, 1932.**

Siegl sent equal parts of identical blood specimens to Dr. Loewenstein and Dr. Maresch for cultivation of tubercle bacilli by Loewenstein's method. A third aliquot portion was sedimented and injected into guinea-pigs. The specimens were obtained from 140 children; some of these had active tuberculosis, others reacted positively to tuberculin but had clinically inactive tuberculosis, and the remainder gave negative reactions to tuberculin. Dr. Loewenstein's laboratory reported 13.5 per cent positive cultures for those with active tuberculosis, 18.9 per cent positive results for those with inactive tuberculosis, and 23.9 per cent positive findings for those with negative tuberculin tests. Dr. Maresch's laboratory reported that 5.1 per cent of the specimens contained tubercle bacilli, and these specimens were all from children with active tuberculosis. Moreover, only once did both these experimenters isolate tubercle bacilli from the same person's blood. Also only one guinea-pig presented tuberculosis following inoculation and that from a specimen which both laboratories had reported as negative. Siegl also distributed urine, sputum and pus containing tubercle bacilli to the same two laboratories, and although many positive results were reported, in only three specimens did both laboratories report identical results. Of the eleven positive cultures that Loewenstein reported for children who gave negative reactions to tuberculin, six produced typical tuberculosis in guinea-pigs. Siegl concludes that it is not as yet possible to resolve these contradictions.

AARON EDWIN MARGULIS.

**BRONCHIOGENIC TUBERCULOSIS IN CHILDREN. L. DE VELASCO, Beitr. z. Klin. d. Tuberk. 81:674, 1932.**

The author investigated the significance of bronchiogenic propagation in the development of pulmonary tuberculosis in childhood. The material consisted of thirty-two children, aged from 9 months to 6 years, who came to autopsy. Thirteen showed undoubted evidence of extension by the bronchiogenic route. In general, these bronchiogenic foci showed the same differentiation into those due to small seedings and those due to coarse seedings as Loeschke has described in adults. In eight cases the source of the propagation was excavation of a primary focus, while in six cases a caseated lymph node had ruptured into a bronchus. In the absence of evidences of hematogenous dissemination, however, caseation of lymph nodes was slight, and intranodular hemorrhagic foci were absent. This likewise parallels the histologic picture of the disease in adults.

AARON EDWIN MARGULIS.

**THE INFLUENCE OF ALTITUDE ON TUBERCULOSIS IN GUINEA-PIGS. LUDWIG LANGE and OTHERS, Beitr. z. klin. d. Tuberk. 82:1, 1933.**

Extensive, identical experiments were conducted simultaneously at four altitudes (sea level, 375 meters, 1,570 meters and 3,100 meters). At each station the animals were divided into two groups, one of which was kept in the dark and the other in diffuse daylight. In addition, a portion of the latter were sensitized to light with eosin. Some of the guinea-pigs were allowed to become acclimated to the altitude before the start of the experiments, and others were inoculated immediately on arrival at the respective stations. All were given injections of similar amounts of identical strains of tubercle bacilli. Records were kept of duration of life and changes in weight, and on autopsy studies were made of the histologic aspects and distribution of the disease. The only significant findings were that the lowest death rate occurred in the nonsensitized animals kept in the light, and the highest death rate in the eosin-sensitized animals kept in the light at high altitudes. No other significant deviations as between the different stations were noted.

AARON EDWIN MARGULIS.

**THE GENESIS OF ACUTE, GENERAL, MILIARY TUBERCULOSIS. I. MINGUEZ,**  
Beitr. z. Klin. d. Tuberk. 82:84, 1933.

This study is based on thirty-nine autopsies. In each Minguez searched for the focus of the spread, using as criteria Loeschke's postulates: liquefaction of contents, presence of a large number of bacilli and presence of blood as an indication of rupture into a blood vessel. He believes that endovascular tubercles are the result and not the cause of bacillemia. In thirty-one cases such foci were certainly demonstrated, and in eight doubtfully so. The focus was in a lymph node in twenty-two instances; it was primary three times; it was in a caseated testicle once, and in the remaining three cases it was indeterminate.

AARON EDWIN MARGULIS.

**EXPERIMENTAL TUBERCULOSIS OF THE BONES. M. MANDELSTAMM,** Beitr. z. Klin. d. Tuberk. 82:98, 1933.

Rabbits were inoculated with bovine tubercle bacilli intra-articularly, intracardially and subcutaneously. Metastatic tubercles appeared in all elements of the bony system except the fat marrow. In general, they were distributed perivascularly, the arteries themselves, however, never showing any tuberculous infiltrations. The tubercles that appeared in the myelogenous bone marrow of the diaphyses did not caseate, while those which appeared in the metaphyses developed more rapidly and frequently caseated. Thus clinical experimental bone tuberculosis in the rabbit, as in the human being, starts in the epiphyseal region. This is interpreted as evidence of significant variation in local resistance of specific tissue.

AARON EDWIN MARGULIS.

**NEW BLOOD VESSEL FORMATION IN PULMONARY TUBERCULOSIS AND ITS SIGNIFICANCE FOR TUBERCLE BACILLEMA. JOSÉ ABÉLLO PASCUAL,** Beitr. z. Klin. d. Tuberk. 82:152, 1933.

Studying sections stained with hematoxylin alum and acid fuchsin, Pascual demonstrated that the new-formed capillaries in richly vascularized tuberculous granulation tissues frequently contain either phagocytosed or free tubercle bacilli. This manner of invading the blood, although not the only one, may have major significance in the development of tuberculosis. AARON EDWIN MARGULIS.

**SERUM LIPASE CHANGES IN EXPERIMENTAL TUBERCULOSIS. J. BALÓ, F. GERLEI and L. LOVAS,** Beitr. z. Klin. d. Tuberk. 82:164, 1933.

Forty rabbits were inoculated with bovine tubercle bacilli. Half received, in addition, intravenous injections of pancreatic extract, at first daily and subsequently at longer intervals. The controls uniformly showed a decrease in serum lipase, while those receiving the pancreatic extract showed either no significant variations or, as happened in most instances, a distinct rise in the serum lipase.

AARON EDWIN MARGULIS.

**TOXIC GRANULATIONS IN LEUKOCYTES IN TUBERCULOSIS. J. LEITNER and W. EICHORN,** Beitr. z. Klin. d. Tuberk. 82:173, 1933.

The authors discuss the significance of toxic granulations in leukocytes. As regards tuberculosis, although the granulations occur most often in the presence of exudative processes, in which sometimes all the polymorphonuclear cells show such granulations, they also occur when the processes are productive. Hence, although they tend to occur in greater number in severe cases, their diagnostic and prognostic value is limited. However, taken in connection with a Schilling or Arneth count, their presence is worthy of consideration.

AARON EDWIN MARGULIS.

**TYPHOID BACILLUS MENINGITIS.** J. WEEBER, Beitr. z. path. Anat. u. z. allg. Path. **92**:223, 1933.

In the Szent-László Hospital of Budapest, Hungary, there were in the years 1922 to 1932, inclusive, 10,327 necropsies. This number included 1,131 cases of typhoid, one of which was complicated by meningitis due to the typhoid bacillus. In this case relapse occurred in the eighth week, and meningeal symptoms developed in the twelfth week, death occurring on the tenth day after the onset of meningeal symptoms. Typhoid bacilli were cultivated from the purulent spinal fluid withdrawn during life. Histologically the process was characterized by a predominance of lymphocytes and by the presence of many large phagocytic cells.

O. T. SCHULTZ.

**YELLOW FEVER VIRUS IN TISSUE CULTURE WITH ESPECIAL REFERENCE TO ITS CULTIVABILITY.** E. HAAGEN and M. THEILER, Zentralbl. f. Bakt. (Abt. 1) **125**:145, 1932.

The virus of yellow fever (French strain) behaves in tissue cultures in general like the viruses of variola, vaccinia, herpes and foot and mouth disease. By Theiler's method of intracerebral injection into mice it was found that the virus requires living cells for its maintenance in vitro; in the absence of living cells or on the addition of killed cells the virulence is soon lost. The virulence in numerous passages remained constant, and the authors therefore conclude that the cultivation of the virus was achieved.

PAUL R. CANNON.

**THE SIGNIFICANCE OF BACTERIUM GRANULOSIS (NOGUCHI) IN TRACHOMA.** C. J. SCHUURMAN, Zentralbl. f. Bakt. (Abt. 1) **125**:158, 1932.

Schuurman made a search for Bact. granulosis in 33 patients with trachoma, using the technic given by Noguchi. Seventy-three strains of bacteria were isolated, none of which was identical with either of two strains of Bact. granulosis obtained from America. He concludes that this organism is of no significance in the etiology of trachoma in Java, and suggests that more attention should be paid to the cell inclusions described by Prowazek.

PAUL R. CANNON.

**EXPERIMENTAL SYPHILIS OF THE CENTRAL NERVOUS SYSTEM OF RABBITS.** T. TANI and K. SAITO, Zentralbl. f. Bakt. (Abt. 1) **125**:417, 1932.

Syphilitic testicular emulsions containing spirochete strain "VIII" were injected into the brains of rabbits to determine the optimum method for obtaining infection, as revealed by the development of a positive Wassermann reaction in the spinal fluid. The best results were obtained in full-grown male albino rabbits receiving injections into the parietal lobes or into the subarachnoidal space with 0.5 cc. of the emulsion, given four times at intervals of one week. Of the sixty-four rabbits given injections, forty-eight lived more than thirty days, and forty-six of these gave a positive Wassermann reaction of the spinal fluid.

PAUL R. CANNON.

### Immunology

**THE TOXIC PROPERTIES OF SERUM EXTRACTS OF HEMOLYTIC STREPTOCOCCI.** J. T. WELD, J. Exper. Med. **59**:83, 1934.

A method is described whereby toxic substances may be extracted from hemolytic streptococci with inactivated serum. Such extracts contain large amounts of hemotoxin and leukocidin. Their intravenous injection into mice causes marked hemoglobinuria, anemia and death. There is evidence that the anemia is not the only cause of death. Incomplete work seems to indicate that the hemotoxin and the lethal poisons are not antigenic. Certain biologic properties of the extract are described.

AUTHOR'S CONCLUSIONS.

TISSUE REACTIONS IN IMMUNITY. REUBEN L. KAHN, J. Immunol. 25:295, 307, 317, 331, 339, 347 and 363, 1933.

The report consists of seven articles. The skin tests for the estimation of the sensitivity of rabbits were done by injecting intradermally 0.1 cc. of increasing dilutions of the antigen. The highest dilution producing an inflammatory reaction after twenty-four hours gave the titer of skin sensitivity. A symbolic formula was employed to express quantitatively the extent and intensity of the inflammatory response (for instance, 4R<sub>3</sub> In<sub>2</sub>, 3B<sub>3</sub> means that the response was a markedly raised [edematous] red area, 4 cm. in diameter with a central black necrotic area 3 cm. in diameter). The titer of precipitins in the serum was determined by mixing 0.1 cc. quantities of dilutions of the antigen with similar quantities of the serum. The tests were read on the following day. There was some correlation between the titers of skin sensitivity and those of the serum precipitins during the first weeks following sensitization; later the precipitins in the serum tended to decrease and eventually to disappear, while the cutaneous sensitivity persisted. Tissue hypersensitivity is a more permanent response than precipitin production, and the two phenomena are independent of one another. Desensitization of rabbits sensitized with human serum was accomplished by intravenous injections of 5 cc. of the serum per kilogram of body weight in one series and 0.5 cc. per kilogram of body weight in another series of four animals with low precipitins or absence of precipitins. The skin sensitivity and the precipitins disappeared following the intravenous injection. They returned gradually after forty-eight hours, the precipitins coming back more rapidly. Desensitization was also successful in rabbits sensitized to egg-white and to horse serum. Heretofore, desensitization was considered possible only in the case of a simple protein in crystalline form. A marked increase in the intensity of the cutaneous inflammatory reactions remaining from previous skin tests was noted following the desensitizing injections. In rabbits receiving desensitizing intravenous injections of bacterial suspensions, necrotic lesions appeared rapidly even in areas where before the intravenous injection no local response was noticeable after intracutaneous injections of the antigen. Intravenous injections of typhoid and paratyphoid A bacilli into immunized rabbits (4 cc. per kilogram of body weight of bacterial suspensions containing 3 billion organisms per cubic centimeter) succeeded in marked, though only temporary, reductions of cutaneous sensitivity, without affecting the titer of agglutinins. Similar injections of typhoid, paratyphoid A and colon bacilli and hemolytic streptococci into normal, not artificially sensitized rabbits removed temporarily their normal cutaneous sensitivity to these micro-organisms and led to a similar increase of intensity of the local inflammatory reactions which remained after tests for cutaneous sensitivity. The results of Schultz and Swift were confirmed concerning the development of a more marked skin sensitivity in rabbits given intradermal injections of hemolytic streptococci, while intravenous injections stimulated mainly a rise of the agglutinin and a relatively weaker cutaneous response. Similar results were obtained with typhoid bacilli. Less frequent injections (every sixteen days) of hemolytic streptococci produced a more marked cutaneous sensitivity than more frequent ones (every four days).

SPECIES SPECIFICITY OF FIBRINOGENS. H. B. KENTON, J. Immunol. 25:461, 1933.

On the basis of anaphylactic reactions in pigeons Kenton concludes that fibrinogens are not of identical structure in all species.

STREPTOCOCCUS LEUCOCIDIN. F. P. GAY and F. ORAM, J. Immunol. 25:501, 1934.

Broth filtrates of a pathogenic hemolytic streptococcus contain a substance, "leukocidin," already described, that causes loss of phagocytic power, visible morphologic changes and loss of the power to reduce methylene blue in leukocytes of certain types and of certain animal species in varying degree.

**ANTIGENIC SUBSTANCES OF CLOSTRIDIUM PARABOTULINUM.** J. B. GUNNISON,  
J. Immunol. 26:17, 1934.

Extracts of toxic cultures of *Clostridium parabotulinum* prepared by the freezing and thawing of young washed cultures contained both protein and carbohydrate, but were not antigenic. These extracts reacted specifically in precipitin tests with antiseraums produced by the inoculation of the intact organisms. An extract prepared from a strain of *Clostridium parabotulinum* type B which had lost its toxicity was antigenic. This extract stimulated the formation of precipitins which were specific for the toxic type, but not for the groups within the type. Two protein substances were obtained from the supernatant fluid of 1 week old broth cultures by precipitation with acid. One of these substances (A), which was soluble in an excess of acid, was antigenic. The other (B), which was insoluble in an excess of acid, was nonantigenic. These substances (A and B) were not type-specific and did not stimulate the production of antitoxin.

FROM THE AUTHOR'S CONCLUSIONS.

**THE DEPRESSION OF PHAGOCYTOSIS BY PRODUCTS OF STAPHYLOCOCCI.** R. M. PIKE, J. Immunol. 26:69, 1934.

Filtrates of staphylococci exert a depressing effect on phagocytosis in vitro. The depressing agent is not antigenic, is thermostable and does not deteriorate on standing. Its production is not affected by an atmosphere containing added carbon dioxide. A similar phagocytosis-depressing substance is produced by the colon bacillus.

FROM THE AUTHOR'S CONCLUSIONS.

**PNEUMOCOCCAL HAEMOLYSIN.** S. T. COWAN, J. Path. & Bact. 38:61, 1934.

Hemolysin is found in the fluid part of young broth cultures of *Pneumococcus*. Hemolysin is not type-specific, and the capacity of a strain to produce hemolysin varies independently of virulence.

**RICKETTSIA VACCINE IN TYPHUS.** RUDOLF WEIGL, Arch. Inst. Pasteur de Tunis 22:315, 1933.

A series of observations were made on persons vaccinated with the phenolized intestines of infected lice. A man, immune to typhus, was employed to feed laboratory lice daily. Without knowledge of Weigl, he gave several hundred to his wife to feed. She had been vaccinated. No attack followed, so several members of the institute repeated the accidental experiment. The results indicated the value of vaccination. One person, vaccinated too late, was attacked but still failed to transmit infection to lice. Thus prophylactic virtues are also claimed for the vaccine, since distribution of the virus through the blood is prevented. The latter observation was confirmed by animal experiment.

M. S. MARSHALL.

**CROSS-IMMUNITY TESTS WITH A RODENT TYPHUS VIRUS FROM ANVERS.** CHARLES NICOLLE and J. LAIGRET, Arch. Inst. Pasteur de Tunis 22:338, 1933.

A virus, isolated from rats from the S. S. Roumanie, which arrived at Anvers March 21, 1933, was tested on guinea-pigs. Cross immunity was manifested against the historic Tunis strain of typhus virus, and against the rodent strains of Mexico and Toulon, but not against the virus of Rocky Mountain spotted fever. Four animals given the latter virus followed by the Anvers strain showed, as a result of the second virus, one clear infection, two light infections and one absence of infection. The viruses were reversed in five animals, resulting in one clear infection, three light ones and one failure or doubtful infection. Since, however, fresh animals usually die from the virus of Rocky Mountain spotted fever, it seemed that the Anvers virus antigenically approached the Montana virus more than other rodent strains thus far studied.

M. S. MARSHALL.

LOUSE FECES FOR RICKETTSIA VACCINE. B. CHRZANOWSKI and H. MOSING, Arch. Inst. Pasteur de Tunis **22**:346, 1933.

The authors prepared a vaccine from the feces of infected lice, purified by fractional centrifugation and washing. From experiments on about 250 guinea-pigs, with the classic Weigl vaccine for control, it was concluded that such vaccine immunized even in small doses (equivalent to the elimination from 2 lice in three days). One gram was sufficient to immunize from 600 to 1,000 animals. No reactions were observed. Feces 2 years old were still effective. It was estimated that the feces of lice during the last three days of infection included two thirds of the number of rickettsiae retained in the intestines during extreme infection. Agglutination occurred with rickettsiae in titers proportional to the amounts of vaccine. It is suggested that this vaccine be added to the intestines (Weigl's vaccine) in the proportion of 1:1.5, thereby reducing the laborious effort required in collecting those organs.

M. S. MARSHALL.

INFLUENCE OF THE VEGETATIVE NERVOUS SYSTEM ON ANAPHYLAXIS. G. GUASSARDO, Pathologica **25**:642, 1933.

Choline and ergotamine, which stimulate the vagus, increase the anaphylactic shock of guinea-pigs sensitized against horse serum if given during the whole period of anaphylactic incubation. Atropine and epinephrine, which stimulate the sympathetic, produce a contrary effect.

E. VON HAAM.

A STUDY OF STAPHYLOLYSIN. J. FORSSMAN, Biochem. Ztschr. **265**:291, 1933.

Forssman found a marked variation in the properties of filtrates of broth cultures of hemolytic staphylococci. The technic of the preparation of the lysin had some, though only limited, bearing on its hemolytic efficiency. The optimum conditions were: a 4 to 9 day old growth in veal broth containing from 4 to 7.5 per cent peptone and about 0.2 per cent of rabbit or sheep blood and a decreased supply of oxygen through the use of specially constructed stoppers. Guinea-pig blood and dextrose added to the medium were unfavorable. Staphylococci growing on blood agar plates are known to take uniformly the red blood cells of a very large variety of animal species. The in vitro determination of the lytic power of filtrates revealed a marked and rather irregular variation. The red blood cells of guinea-pigs and horses were not laked and those of man only slightly, while those of the goat, sheep, ox and particularly the rabbit were readily hemolyzed. Variations were noted with different specimens of blood from the same individuals. Equally marked variations were caused by different degrees of temperature. The optimum temperature was 37 C. for two hours, followed by twenty hours at 10 C. The red blood cells of rabbits were laked at the end of the first period of incubation, while those of the other species showed then only very slight hemolysis or none, the main part of the lysis occurring during the subsequent incubation at the lower temperature. The laking of red blood cells is the only common feature of immune hemolysins and of staphylysin. The effect of the latter is inversely proportionate to its dilution and directly proportionate to the concentration of the red blood cells, both properties being diametrically opposite to those possessed by immune hemolysins. Forssman's observations on the adsorption of staphylysin by various red blood cells differ from those previously reported. He found no relation between the adsorption and the lysis; in other words, the adsorption occurred even with red cells known to be resistant to lysis and vice versa. The adsorption was irregular. A striking phenomenon was a marked but only temporary adsorption of the lysin at 0 C. by red blood cells of some species. The lysin was liberated into the fluid after very short periods of time. The latter quality, the increase of the lytic potency with an increase of the concentration of red cells, and the possible, though not definitely established, activation by the blood cells suggest that the staphylysin is ferment-like.

I. DAVIDSOHN.

ALTERATION IN THE ANTIBODY FORMATION IN RABBITS. E. HAUSMANN, Frankfurt. Ztschr. f. Path. 45:431, 1933.

Precipitins, agglutinins and hemolysins were produced in rabbits by injection of foreign serum and red blood cells respectively. While injections of epinephrine and pilocarpine did not cause any alteration in the production of antibodies, a definite increase in the formation of all the three antibodies was produced by the injection of atropine.

W. SAPHIR.

THE MODE OF ACTION OF IMMUNE SERUM IN SWINE ERYSIPelas. DEZSÖ GAJZÁGÓ, Zentralbl. f. Bakt. (Abt. 1) 125:89, 1932.

Gajzágó studied the effects of passive immunization of mice and pigeons with an antiserum for *Erysipelothrix rhusiopathiae*. He observed no bactericidal action and found no injurious effects on the micro-organisms. Phagocytes played the predominant rôle, although bacteriotropins were not exclusively concerned, inasmuch as serums in which bacteriotropins had been removed by saturation still were protective. Bacteria injected intraperitoneally into immune animals agglutinated in the peritoneal cavity and were more effectively phagocytosed than in normal animals. Furthermore, bacteria could be recovered from the blood stream of the normal animal for from one to two days, whereas in the immune animal the micro-organisms persisted in the blood stream only to the third hour.

PAUL R. CANNON.

THE EFFECT OF "BLOCKADE" OF THE RETICULO-ENDOTHELIAL SYSTEM ON THE COURSE OF VACCINIA. ALEXANDER HASSKÓ, Zentralbl. f. Bakt. (Abt. 1) 125:166, 1932.

Rabbits were given intravenous injections of a 10 per cent solution of saccharated iron and after several weeks' treatment were infected via the scarified skin with raw vaccinal lymph, and the effects of generalization of the virus were compared with those observed in normal rabbits similarly affected. The virus generalized much more markedly in the "blockaded" animals, and the infection was much more severe. Many of the animals died from secondary infections. No quantitative differences in the concentration of virus in the blood of normal and "blockaded" rabbits were demonstrable, however, by inoculation of the blood into the scarified skin of guinea-pigs. Experiments on the effects of generalization of the virus in normal and splenectomized guinea-pigs, cutaneously infected, show a more marked generalization in the latter.

PAUL R. CANNON.

THE BORDET-WASSERMANN REACTION IN TYPHUS FEVER. L. HINSZELD and W. HALBER, Acta path. et microbiol. Scandinav., supp. 16, p. 114, 1933.

The Wassermann reaction is not absolutely specific for syphilis. It may be positive in typhus, leprosy, trypanosomiasis, malaria, endocarditis lenta, measles and the last stages of cancer. A comparison was made of Wassermann reactions in syphilis and in typhus, using various antigens and absorption tests with lecithin. In syphilis there is a reagent against organ extracts. In typhus this could not be demonstrated. The theory of these phenomena is discussed.

JACOB KLEIN.

RELATION BETWEEN ALLERGY AND IMMUNITY IN TUBERCULOSIS. OLUF THOMSEN and K. PEDERSEN-BJERGAARD, Acta path. et microbiol. Scandinav., supp. 16, p. 521, 1933.

This is an attempt to solve experimentally the question whether allergy in the presence of tuberculous infection increases immunity and favorably influences resistance against superinfection as well as against the existing infection. A series of guinea-pigs was sensitized to horse serum. An emulsion of tubercle

bacilli in horse serum was then injected, and the results were compared with those in nonsensitized controls. The local anaphylactic reaction had no influence on the tuberculous process. Nor was there any favorable influence on superinfection.

JACOB KLEIN.

THE LEUKOCYTES IN ISO-AGGLUTINATION. IVAR WALLGREN, Acta path. et microbiol. Scandinav., supp. 16, p. 556, 1933.

Agglutination was carried out with whole blood in Tyrode's solution and 25 per cent serum. The preparations were examined in a warmed chamber with the dark field. Clumps of erythrocytes were found about leukocytes as a center. This did not seem to interfere with the viability of the leukocytes. When the agglutination ceased, the erythrocytes were taken up by the leukocytes. No agglutination of the leukocytes took place. In normal blood under these conditions auto-erythrophagocytosis takes place. There are serums which stimulate this process. B-serum and A-B serum induced phagocytosis of B-erythrocytes and also O-erythrocytes. This would not happen were the isoantibodies group-specific. The agglutinins and the opsonins are not identical.

JACOB KLEIN.

### Tumors

EFFECTS OF THYROIDECTOMY ON TUMOR GROWTH IN MICE. M. LEVINE and V. H. KUGEL, Am. J. Cancer **19**:817, 1933.

The general tendency seemed to be for the thyroid-injured mouse to show smaller tumors than the control although the weight of the animal was favored by the thyroid injury.

AMYLOIDOSIS IN MULTIPLE MYELOMA. O. S. RANDALL, Am. J. Cancer **19**:838, 1933.

A case of multiple myeloma of bone is reported in which amyloid infiltration of the small intestine resulted in intestinal obstruction. Although amyloidosis of the intestine is rare as a complication of multiple myeloma, one should at least suspect it when complaints referable to the gastro-intestinal tract are made. Inasmuch as gastro-intestinal symptoms are reported in 20 per cent of all cases of multiple myeloma (Geschickter and Copeland), amyloidosis of the intestine may be a more frequent complication than is generally believed. Amyloidosis should be considered in every wasting and long-standing illness, whether or not suppuration is present.

FAILURE OF ETHYL ALCOHOL IN MOUSE CANCER. M. C. MARSH, Am. J. Cancer **19**:847, 1933.

Ethyl alcohol was administered in various concentrations, at maximum dosage, over long periods, by intravenous and subcutaneous injections, by ingestion and by combinations of these, to forty-nine mice with spontaneous tumors. There was no evidence of any curative effect.

AUTHOR'S SUMMARY.

METAPLASTIC AND NEOPLASTIC BONE- AND CARTILAGE-CONTAINING TUMORS OF SOFT PARTS. T. B. MALLORY, Am. J. Path. **9**:765, 1933.

Many apparent bone-containing and cartilage-containing tumors of soft parts are in reality of metaplastic rather than neoplastic origin. Extreme care may be requisite to differentiate the active stage of osteoblastic or chondroblastic metaplasia from malignant neoplasia. Certain bone-forming spindle cell sarcomas of soft parts may more logically be regarded as fibrosarcomas with metaplastic foci of osteogenesis than as true osteoblastomas.

AUTHOR'S CONCLUSIONS.

**CONGENITAL LYMPHOBLASTOMA.** C. F. BRANCH, Am. J. Path. 9:777, 1933.

The literature concerning cases of congenital lymphoblastoma is reviewed. A case of congenital lymphoblastoma is presented in which the normal architecture of the organs had been virtually destroyed by a massive lymphocytic infiltration.

**AUTHOR'S SUMMARY.****THE ANGLE OF THE MITOTIC SPINDLES IN MALIGNANT CELLS.** S. WARREN, Am. J. Path. 9:781, 1933.

On the basis of the cases thus far studied the angle of the mitotic spindle provides a criterion by which carcinoma can be distinguished from sarcoma and from lymphoblastoma. Because of the two types of mitosis apparently usually occurring in the same tissue in fibrosarcoma, as distinct from other types of sarcoma, this particular form may be distinguished. The mean values obtained for the mitotic angles are: in carcinoma, 91 degrees; in melanotic sarcoma, 56 degrees; in fibrosarcoma, 72 degrees for the narrow spindles and 108 degrees for the wide spindles; in osteogenic sarcoma, 69 degrees, and in Ewing's tumor, 88 degrees. Further study of other types of tumors and normal cells is now in progress.

**AUTHOR'S SUMMARY AND CONCLUSIONS.****THE EFFECT OF CAESIUM CHLORIDE ON TRANSPLANTED TUMORS OF MICE.** A. W. WRIGHT and C. F. GRAHAM, Am. J. Path. 9:789, 1933.

The hypothesis that the introduction of strongly basic ions into the nuclear complex might greatly reduce nuclear activity is not supported by the study of the effect of cesium on the growth of the Twort carcinoma. There is no evidence that it affects in any way the capacity of the tumor cells to divide and multiply. There is, however, suggestive evidence to indicate that necrosis of the neoplastic tissue may be more extensive in the cesium-treated animals. That cesium under certain conditions has a deterrent effect on the growth of the tumor is indicated, and further studies of this property of the element are in progress.

**AUTHOR'S SUMMARY AND CONCLUSIONS.****THE DIAGNOSIS OF TUMORS BY ASPIRATION.** F. W. STEWART, Am. J. Path. 9:801, 1933.

Needle aspiration is an expeditious practical method for the diagnosis of tumors. In approximately twenty-five hundred cases my associates and I have observed no untoward result following its use. The interpretation of smears of aspirated material often requires competent clinical assistance. Diagnosis by aspiration is as reliable as the combined intelligence of the clinician and the pathologist makes it. The pathologist who ventures to interpret the material obtained by aspiration will have to revise or relearn many criteria. The clinician must appreciate how far the pathologist can logically go in interpreting the smear. Both must maintain a sympathetic attitude toward a new procedure. In the institution with which I am associated the method has so established its usefulness that it has acquired a permanent place as a means of diagnosis.

**AUTHOR'S SUMMARY.****PRECANCEROUS EPIDERMAL LESIONS.** M. B. SULZBERGER and D. L. SATENSTEIN, Arch. Dermat. & Syph. 28:798, 1933.

A man, aged 39, had had a lesion on the glans penis for three months. It continued to spread. The lesion consisted of erythematous plaques from which tiny droplets of clear fluid exuded. Biopsy showed hyperplastic epidermis, intra-cellular and intercellular edema and vacuolation. The superficial dermis showed vascular dilatation and marked infiltration by round cells, plasma cells and

eosinophils. The lesion should be considered as precancerous and treatment should be destructive. In dermatologic literature this condition is called erythroplasia of Queyrat.

S. W. BECKER.

**TUMORS OF THE SYMPATHETIC NERVOUS SYSTEM: NEUROBLASTOMA, PARAGANGLIOMA, GANGLIONEUROMA.** D. LEWIS and C. F. GESCHICKTER, Arch. Surg. **28**:16, 1934.

These three types of tumors are regarded as developing from cells which wander out from the neural crest during embryonic life. The undifferentiated cell may give rise to neuroblastoma and the more differentiated to paraganglioma and ganglioneuroma. All three types of growth in the same tumor indicate a common origin.

**LYMPHANGIO-ENDOTHELIOMA OF BONE.** D. A. DESANTO, Arch. Surg. **28**:66, 1934.

A tumor is described which is traced to the perivascular lymphatics of the haversian canals and subperiosteal tissue. This tumor fulfills every criterion of the so-called Ewing's tumor of bone.

**THE AGENTS IN ANILINE CARCINOMA OF THE BLADDER.** G. H. GEHRMANN, J. Urol. **31**:126, 1934.

Tumor of the bladder occurs more frequently among dye workers than among workers not in contact with dyes, and in my experience it occurs to the extent of 4.5 per cent. The combined experiences of the dye industries of Germany, Switzerland, England and America indicate that probably aniline, *alpha* and *beta* naphthylamine and benzidine are the etiologic compounds. Tumor of the bladder may appear any time after two years' exposure, and removal from the dye operation does not eliminate the possibility of future developments. The adoption of a completely closed and properly ventilated process, together with careful medical supervision and protection, will eliminate the hazard, but these methods will require many years of active enforcement and must continue as long as the compounds are made.

FROM AUTHOR'S SUMMARY.

**GENESIS OF ANILINE CARCINOMA OF THE BLADDER.** D. M. GAY, J. Urol. **31**:137, 1934.

Whether the injurious agent exerts its first effect on the submucous blood vessels or directly on the epithelium cannot be proved with the material at hand; but I hold to the hypothesis that aniline tumors are caused by an agent to which the tissues of the bladder are specifically vulnerable. Although the greatest changes are in the region of the trigon, the multiplicity and subsequent origin of tumors in different sites lead me to believe that the injury involves the entire bladder, probably owing to the circulation of a cancerogenic agent in the blood.

FROM AUTHOR'S SUMMARY.

**THE EFFECT OF HYPOPHYSECTOMY ON THE GROWTH OF THE WALKER RAT TUMOUR.** C. S. MC EUEN and D. L. THOMSON, Brit. J. Exper. Path. **14**:384, 1933.

The growth of the Walker rat tumor in the young male albino rat is retarded by hypophysectomy, and the resistance of the animal is lowered. Metastases occur less frequently in hypophysectomized rats, probably because large tumors are rare. Inoculation into hypophysectomized rats is successful in a large proportion of cases, but the growth of the transplants is slow. The total weight of the hypophysectomized tumor-bearing rat remains constant but that of the control is increased. If this increase is prevented by restricting the intake of food, the growth

of the tumor is retarded. This retardation is not significantly less than that produced by hypophysectomy. The body weight of the rat minus the tumor tends to decrease; this decrease is approximately equal to the weight of the tumor in the hypophysectomized rat, but not in the normal rat.

AUTHORS' SUMMARY.

THE REACTION OF THE ARTERIAL BLOOD IN CANCER. SYLVIA DICKINSON and R. E. HAVARD, Brit. J. Exper. Path. **14**:394, 1933.

The results of previous workers are summarized and discussed. The technic is described for collection of samples of arterial blood from patients with and without malignant diseases, and for measurement of the  $p_{H_2}$  of the blood at 37 C. by two independent glass electrodes. The results support the conclusion that the blood of patients with malignant disease of the mouth or skin, or with secondary conditions of the glands, is not more alkaline than that of patients free from malignant disease. An experiment is described which demonstrated that momentary alterations of respiration can affect the  $p_{H_2}$  of the blood.

AUTHORS' SUMMARY.

ON THE NATURE OF THE TUMOURS INDUCED IN FOWLS BY INJECTIONS OF TAR. JAMES MCINTOSH, Brit. J. Exper. Path. **14**:422, 1933.

Tumors of sarcomatous type can readily be induced in the fowl by intramuscular injections of tar and lard. Four tar-induced tumors have been transmitted in series in fowls by injections of tumor emulsion, and three of these have been transmitted by means of cell-free filtrates (Berkefeld). Tar-induced sarcomatous tumors of the fowl conform closely in morphologic and biologic characteristics with the naturally occurring sarcomatous tumors of the fowl.

AUTHOR'S SUMMARY.

BONE FORMATION IN CARCINOMA OF THE GALLBLADDER AND IN ITS METASTASES. G. MICSEK, Frankfurt. Ztschr. f. Path. **44**:430, 1933.

The primary carcinoma of the gallbladder as well as metastases in the lungs, liver and lymph nodes showed typical bone and bone marrow formation. The presence of cancer cells apparently stimulated proliferation of connective tissue. The newly formed mesenchymal cells were then secondarily transformed into bone and bone marrow cells as the result of metaplasia due to a specific stimulation of the carcinoma cells.

WILLIAM SAPHIR.

THE RELATION OF THYROID ADENOMA AND GOITER TO THE CAPSULE OF THE THYROID AND TO ITS BLOOD VESSELS. F. DOEPFNER, Frankfurt. Ztschr. f. Path. **44**:461, 1933.

Four instances of so-called metastasizing thyroid adenoma, seventeen instances of simple thyroid adenoma and four of proliferating goiter have been studied. In nearly all cases an invasion into the connective tissue capsule of the thyroid gland was noted. In the four cases of metastasizing adenoma, in three of the instances of simple adenoma and in three of the cases of proliferating goiter tumor tissue had broken into the veins. These observations lead Doepfner to believe that invasion of tumor tissue into blood vessels does not always of itself lead to metastases, but that certain other factors, such as age of the patient, play an important rôle in the production of metastases.

WILLIAM SAPHIR.

MYELOID CHANGES IN THE SPLEEN OF MICE TREATED WITH TAR. N. WATERMAN, Frankfurt. Ztschr. f. Path. **44**:540, 1933.

Waterman calls attention to the myeloid transformation of the spleen regularly found in the mouse with tar carcinoma. Since this change is never observed in the mouse with tar papilloma it is assumed that tar cancer can develop only when myeloid changes in the spleen have taken place.

WILLIAM SAPHIR.

## Medicolegal Pathology

OXIDATION OF ALCOHOL WITH RELATION TO MEALS. GOTTFRIED JUNGMIEHEL, Deutsche Ztschr. f. d. ges. gerichtl. Med. 22:11, 1933.

If alcohol is ingested on a fasting stomach there is a greater concentration in the blood after drinking pure diluted alcohol than after drinking beer. With a previous intake of food there is a diminution of alcohol in the blood due to the altered conditions of absorption from the gastro-intestinal tract. A moderate caloric intake is associated with a greater oxidation of alcohol than an excessive intake of food.

JACOB KLEIN.

EXPERIMENTAL STUDIES ON DEATH FROM DROWNING. NICOLAE P. BALAN. Deutsche Ztschr. f. d. ges. gerichtl. Med. 22:167, 1933.

Brouardel distinguishes five phases in the act of drowning: (1) excitement, (2) apnea, (3) deep inspiration, (4) asphyxia and (5) terminal inspirations. Some investigators doubt that water enters the lungs in drowning, and state that death is due to pulmonary edema. To clarify this question experiments were done on twenty guinea-pigs in water containing a suspension of rice flour, which is readily seen under the microscope. After drowning the lungs were removed and studied. The rice granules were not found in the lung until the third or dyspneic stage. The water first entered the upper part of the lung; intensive inspiration drew it into the lower lobes. Previous views are corroborated that water enters the respiratory passages and lungs during drowning, especially in the period of dyspnea, and that death is due to the resulting asphyxia.

JACOB KLEIN.

OBSERVATIONS ON EXECUTION BY HANGING. E. KALLE, Deutsche Ztschr. f. d. ges. gerichtl. Med. 22:192, 1933.

Observations on twenty-four persons executed by hanging indicate that death does not occur suddenly; that pulse and cardiac activity may be noted even twenty minutes afterward. The strangulation causes severe injuries to the structures in the neck. Fracture or luxation of the cervical vertebrae is most rare. Reflex muscular activity is frequently observed and not to be ascribed to conscious movement. The period of deepest unconsciousness occurs at the moment of the drop when the loop begins to act.

JACOB KLEIN.

ROENTGEN DEMONSTRATION OF A METAL RING AT THE POINT OF ENTRANCE OF A BULLET. L. M. EIDLIN, Deutsche Ztschr. f. d. ges. gerichtl. Med. 22:204, 1933.

Roentgen examination demonstrates a lead ring at the point of entrance of a lead bullet. With a steel jacket bullet the metal ring is present in the skin with a shooting distance of 25 to 40 mm. The metal ring aids in distinguishing between the points of entrance and exit of the bullet. Contamination of the wound with blood, organic materials and cleaning of the clothing does not interfere with the demonstration. This marking may be the only available information as to the type of weapon, the distance of the weapon and the direction of the shot. The metal ring is less definite with a wound from a cleaned or a new weapon than with a used or an unclean one. The angle of inclination of the weapon may also be determined in this way. The advantages of this method are: simplicity, availability, noninterference with other examinations and accuracy.

JACOB KLEIN.

PERFORATION OF THE DRUM MEMBRANE AND DEATH FROM DROWNING. M. MILOVANOVIC, Deutsche Ztschr. f. d. ges. gerichtl. Med. 22:427, 1934.

A thorough examination of sixty-seven cases of drowning did not yield any evidence supporting the view of a causal connection between perforation of the drum membrane and death from drowning.

ACUTE VERONAL POISONING. S. SCHEIDECKTER, Deutsche Ztschr. f. d. ges. gerichtl. Med. 22:452, 1934.

In two cases of death from acute veronal poisoning distinct changes due to anemia appeared in the ganglion cells a few hours after the poison was taken. The lipoid changes in the ganglion cells and glia were pronounced. A smaller degree of lipoid accumulation was observed also in the endothelial cells of other organs and in the Kupffer cells.

ELECTRIC INJURY AND DEATH. S. KOEPPEN, Virchows Arch. f. path. Anat. 290:460, 1933.

A young electric worker who, in the course of his work, had been subjected to repeated minor electric shocks that caused no immediate damage began to have symptoms of multiple sclerosis and half a year later died. Autopsy revealed multiple older areas of softening in the brain and recent thrombosis of the sigmoid sinus. Nothing could be found that might explain these lesions. This case raised questions relative to late injury by the electric current. These, as well as controversial questions on the immediate injury caused by electricity and on the mechanism of death, were subjected to experimental investigation. Guinea-pigs, rabbits and dogs were used. The direct current was used, at a strength of 20 to 70 volts in smaller animals and up to 220 volts in larger animals. Single shocks and repeated shocks of one to thirty seconds' duration were applied. In some animals the current was applied at intervals of a month with the aim of producing late changes; the results of such experiments are not included in this report. The positive electrode was usually placed on the head, the negative one on the thigh. Some animals died as the result of the shock and were subjected to immediate gross and microscopic examination. Others that did not die were killed after application of the current and examined. Still others were permitted to live and were killed at varying intervals. Koeppen recognizes local and general effects of the current. The local effects were edema, loss of hair, necrosis, ulceration and infection. Edema and loss of hair are the result of vascular reactions, such as predominate in the general effects of the current. Necrosis and the formation of spaces in the epithelium are specific electric effects due to electrolysis; they are not due to burning or heat; the heat generated in the tissue by the weak currents used was measured and found to be within the limits of the normal production of heat by the tissue. Infection is secondary to death of tissue. The chief and essential general or distant effect is the vascular reaction. This is characterized by extreme distention of the capillaries and veins, with hyaline and red cell thrombosis and diapedesis of red corpuscles. The arteries are contracted. This vascular reaction Koeppen interprets, in accordance with the theories of Ricker, as a stasis paralysis of the capillary bed and a spasmodic contraction of the arterial system which is carried back to the heart. The coagulation time of the blood is decreased, and the viscosity is increased. The cerebral effects, which manifest themselves as an epileptiform seizure with loss of consciousness, are due not to the direct action of the current on the brain, but to the vascular reaction of the cerebral vessels, which is the same here as in the other organs. This contention is proved by the fact that animals decerebrated above the thalamus, in which there was no cerebrum to be directly affected by the current, presented exactly the same manifestations as those with intact brains. Koeppen concludes that the local effects of the electric current are specific and are due to electrolysis. They may be complicated or overshadowed by burns if the current is strong enough or if arcing occurs. He maintains that death is due, not to the effect of the current on the brain or to asphyxia, as Jellinek, Schridde and others have claimed, but to the cardiovascular reaction induced by the current. The therapeusis of electric shock therefore requires not only artificial respiration, but agents that will relieve the arterial spasm.

O. T. SCHULTZ.

PREGNANCY AND RUPTURE OF THE AORTA. H. UEBERMUTH, Zentralbl. f. Gynäk. 57:1633, 1933.

Including the case reported in this article there appear to be recorded in the literature only eight cases of rupture of the aorta in pregnancy. The ruptures occurred at the end of pregnancy on the basis of atherosclerotic or syphilitic changes in the vessel. It is pointed out that aneurysm of the aorta is a clearcut indication for the interruption of pregnancy.

THE USE OF ALCOHOLIC SUSPENSIONS OF SPIROCHAETA PALLIDA FOR THE SEROLOGIC DIAGNOSIS OF SYPHILIS. R. R. HOELTZER, Ztschr. f. Immunitätsforsch u. exper. Therap. 80:368, 1933.

Spirochaeta pallida was suspended in a solution of alcohol in physiologic solution of sodium chloride. The suspension was used as an antigen for the complement fixation test for syphilis. It did not become anticomplementary during a period of twelve months, and it became only moderately less sensitive. In a comparative study with other methods the Kahn test proved more sensitive but slightly less specific, while all other antigens were less sensitive than the one prepared from spirochetes.

I. DAVIDSOHN.

NITROBENZENE POISONING. GUNNAR AHLGREN, Acta path. et microbiol. Scandinav., supp. 16, p. 6, 1933.

A medical student died four and one-half hours after the ingestion of nitrobenzene with suicidal intent. Death was due to respiratory failure. The oxygen capacity of the blood was 7.8 per cent one-half hour before death. The urine withdrawn from the bladder after death contained no albumin, reducing substance, blood pigment or sediment. Autopsy was not done. Experiments on dogs indicated an early anuria, although the kidney showed no histologic change. This would explain the finding of normal urine. The clear urine had been excreted before the ingestion of nitrobenzene, which had rapidly caused anuria.

JACOB KLEIN.

THE BLOOD GROUP QUALITIES IN HUMAN URINE AND THEIR CONCENTRATION AND QUANTITATIVE DETERMINATION. ERIK JORPES and GUNNAR NORLIN, Acta path. et microbiol. Scandinav. 11:91, 1934.

The urine was concentrated by evaporation, dialysis and precipitation with alcohol; 120 Gm. of dry water-soluble substance was obtained from 180 liters of A urine and 155 Gm. from 240 liters of B urine. The quantities of the A or B substance in such concentrates before and after various procedures were determined by adding them in watery solution to human iso-agglutinating serums and by subsequently determining the decrease of the iso-agglutinating properties of such serums. The blood group qualities proved highly heat-resistant (no change after one-half hour at 100 C.). They were also able to resist the action of acids, but were quickly destroyed by alkalis. Jorpes and Norlin interpret the latter finding as speaking against the hypothesis of a polysaccharide nature of the blood group substances, for polysaccharides are known to be particularly resistant to alkalis, while a racemization of proteins by alkalis destroys their antigenic properties.

I. DAVIDSOHN.

# Society Transactions

## CHICAGO PATHOLOGICAL SOCIETY

*Regular Monthly Meeting, April 9, 1934*

*E. H. HATTON, President, in the Chair*

### THE SOURCE OF CALCIUM CARBONATE IN GALLSTONES. D. B. PHEMISTER.

The three principal constituents of gallstones are cholesterol, bile pigment and calcium carbonate. These substances may be present singly or in combinations giving a variety of stones. The pigment is derived from bile. There has been much difference of opinion as to the source of the calcium carbonate and the cholesterol. Some claim that they come from the bile, and others, that they are secreted by the wall of the gallbladder. A series of cases of gallstones and experiments on animals have shown that calcium carbonate is thrown out into the gallbladder when the cystic duct or ampulla is obstructed. In eighteen cases of either marked or complete obstruction by an ordinary gallstone calcium carbonate was precipitated in the gallbladder in amounts varying from 0.5 to 20 Gm. It was present as a paste mixed with mucus, as a coarse sand or as large white stones in the most marked cases. In some cases the calcium carbonate incorporated other stones. The surface deposits on stones in the gallbladder have also been found to be laid down in the presence of duct obstruction. Additional evidence that the layers of calcium carbonate in mixed stones are associated with duct obstruction is obtained from cholecystography. When the gallstones cast ring shadows in x-ray pictures owing to the presence of calcium, the gallbladder does not become visualized with cholecystography. Nuclear deposits of calcium in gallstones are more difficult to explain, and there is greater uncertainty as to the source of the calcium. An inflammation in the gallbladder interfering with free filling and with concentration of the bile may be responsible for the precipitation of calcium forming nuclei on which subsequent deposits of pigment and cholesterol are made.

#### DISCUSSION

E. F. HIRSCH: When the cystic duct is ligated, is there a shift in the reaction of the fluid contained in the gallbladder?

D. B. PHEMISTER: Yes, there is an increase in the alkalinity.

### GENERALIZED ANGIOSARCOMA OF THE LYMPH NODES. PERRY J. MELNICK.

An unusual case of generalized angiosarcoma of the lymph nodes of eighteen months' duration in a 64 year old white man was reported. Biopsies of four different lymph nodes revealed the same structure, namely, an angiofibromatous tumor of low malignancy, centrally located in the lymph nodes, fairly well encapsulated, and appearing independent of the lymphatic tissue or the reticulo-endothelial tissues. The origin seemed to be from multiple foci of angiofibroma in the lymph nodes. The relationship to other forms of multiple angiomas and to Kaposi's multiple angiosarcomatosis of the skin was discussed. Three similar cases of generalized sarcomatosis of the lymph nodes have been reported previously.

#### DISCUSSION

P. DELANEY: The presentation is confused because the primary focus, whether hemangiomatous or lymphangiomatous, is not disclosed.

P. J. MELNICK: The comments by Dr. Delaney are true, for the material is entirely surgical and there has been no postmortem examination.

**SIMULTANEOUS OCCURRENCE OF TWO TUMORS (GLIOMA AND SARCOMA) IN THE CEREBRAL HEMISPHERE OF A CHILD. PERCIVAL BAILEY and ADOLFO LEY.**

We have found only two accounts of the simultaneous occurrence of glioma and sarcoma in the same cerebrum, one by Merzbacher and Uyeda and the other by Wohlwill. The condition seems sufficiently rare to justify a brief report.

Our case occurred in a boy, aged 6 years, who complained of headaches a few weeks before admission to the hospital. He had symptoms of increased intracranial pressure and left spastic hemiparesis. An osteoplastic exploration was made, and a large amount of tumor tissue was removed from the temporal lobe through a transcortical incision. He recovered promptly but acquired otitis media, after which he began to have convulsions, and the site of the operation became tense and bulging. It was reopened and more tumor tissue removed, but the child gradually failed and died.

Aside from the suppuration in the middle ear, necropsy disclosed bilateral fibrous obliterative pleuritis and remnants of tumor in the right temporal lobe of the brain, extending back to the occipital region. In addition there were an extensive ependymitis of all the cerebral ventricles and a small abscess in the old operative wound of the brain.

The tumor removed at the first operation was definitely sarcoma. It was cellular, swarming with mitotic figures. Around the blood vessels were rings of reticulin extending out for some distance into the tissue. The nuclei were vesicular, and in some parts the cells were spindle-shaped with numerous reticulin fibrils between the cells. When the tumor removed at the second operation was examined, we were surprised to find in some portions a structure that was definitely glioma. Here no mitoses were seen, the cells were definitely astroblasts, and the reticulin was confined strictly to the walls of the blood vessels.

At necropsy, in the occipital region, the two tumors could be clearly distinguished. On cross-section, the sarcoma was firm, white and smooth, and the glioma was gray, soft and granular.

The sarcoma must have been of more recent origin, to judge by the numerous mitoses which it contained.

A complete description of the tumors with numerous illustrations will be published in the *Archivos de neurobiología*.

#### DISCUSSION

E. F. HIRSCH: What tissues do you think gave rise to the sarcoma?

P. BAILEY: The sarcoma may have originated from tissues in or about blood vessels.

**INTESTINAL OBSTRUCTION BY A SUBMUCOUS LIPOMA. J. D. KIRSHBAUM.**

In a series of 5,754 autopsies at the Cook County Hospital since 1929 there were 9 lipomas of the gastro-intestinal tract. Two of them were submucous and produced symptoms of intestinal obstruction. The first case revealed at autopsy a sequestration of a mobilized lipoma in the lower ileum causing obstruction and diffuse peritonitis. The second case presented at operation an intussusception of the lower 8 inches (20 Cm.) of the ileum into the cecum. On release of the bowel a submucous lipoma in the ileum was resected. The patient died four days later, and the postmortem examination revealed acute generalized peritonitis. Both patients were ill for only two days: one, a man aged 43 years, and the other, a woman aged 42 years. Lipomas were third in frequency among the 47 benign tumors found in the 5,754 necropsies.

*Regular Monthly Meeting, May 14, 1934*

E. H. HATTON, *President, in the Chair*

THE RÔLE OF THE AVIAN TUBERCLE BACILLUS IN THE ETIOLOGY OF HODGKIN'S DISEASE. PAUL E. STEINER.

Diseased lymph node and splenic tissues from 15 patients with Hodgkin's disease (lymphogranulomatosis) together with tissues from 8 patients with other lymphomas were investigated (1) for the ability of the diseased tissues to produce tuberculosis in chickens, guinea-pigs, rabbits, dogs and mice, and (2) for the disease-producing capacity of these tissues as grafts. Diseased tissues from the 23 patients were injected or transplanted into 199 animals. An additional group of 23 chickens without injections served as controls of environmental factors, especially of spontaneous tuberculosis. A group of 8 dogs received intracerebral injections. The animals survived for periods varying from nine to thirteen months. Criteria for the diagnosis of tuberculosis in the animals were: (1) the acquisition of a positive tuberculin reaction; (2) characteristic gross and microscopic morphologic structures; (3) the presence of stainable acid-fast bacilli in suspicious lesions; (4) the growth of acid-fast bacilli from such lesions on culture mediums; (5) the production of tuberculosis in animals in passage experiments.

In the entire group of animals into which the diseased human tissues were injected, tuberculosis occurred in 1 chicken and 1 guinea-pig given injections of tissue showing Hodgkin's disease and in 1 guinea-pig given an injection of lymphosarcomatous tissue. Animals which received injections of material from tuberculous adenitis became diseased. In the diseased chicken, a spontaneous infection was not excluded. In passage experiments with tissues from the infected animals, tuberculosis was produced.

Numerous lesions in diseased chickens superficially suggesting tuberculosis were considered nontuberculous because they did not satisfy the five criteria mentioned.

No evidence was found that the diseased human tissues were transplantable or that they were capable of inducing lesions with a similar histologic structure in animals.

No strains of acid-fast bacteria were grown by modern cultural methods from tissues of patients with Hodgkin's disease. Likewise the occurrence of acid-fast forms of bacteria reported to exist transiently early in the cultures of such tissues was not confirmed.

Tuberculin protein (T. P. T., Seibert) was used in making intracutaneous skin tests on 35 patients with Hodgkin's disease, and on 38 control patients with a variety of lymphomas. Tuberculin proteins prepared from both avian and human strains were used in comparative quantitative tests for the purpose of obtaining information on the possible etiologic rôle of the avian tubercle bacillus in Hodgkin's disease.

No evidence of specific sensitization to the avian tuberculin protein was obtained by these tests. A marked desensitization (or absence of sensitization) was found to both proteins in the patient with Hodgkin's disease. This decreased "normal adult sensitization" had diagnostic value, especially in differentiating tuberculous adenitis. The incidence of absence of sensitization was greater in the Hodgkin's disease group than in the few with other lymphomas.

These tests suggest that either Hodgkin's disease desensitizes its victims to human and avian tuberculin proteins or it occurs usually in persons who are unable to acquire normal sensitization to these proteins. It is difficult to understand how either of these phenomena could occur in a disease unrelated to tuberculosis.

Within the limits of the adequacy of the testing methods used the avian tubercle bacillus seems to be eliminated as an etiologic factor in Hodgkin's disease. The absence of sensitization to the avian and human tuberculin proteins suggests that

some organism antigenically related to the tubercle bacillus might be etiologically involved. The investigation is being continued with this in mind.

(A complete report of part of this work appeared in the June issue, p. 749.)

#### DISCUSSION

PAUL R. CANNON: Have you noticed a similar decreased sensitization to other substances in patients with Hogkin's disease?

L. E. DAY: It would be interesting to correlate data on the geographic distribution of avian tuberculosis in the United States with similar data on the incidence of Hodgkin's disease. In North America there is no avian tuberculosis, or very little, along the Atlantic Coast. The Middle West has a great deal. If the two diseases are related etiologically, the geographic distribution should be similar. Do you recall whether the lesions observed along the intestinal tract in fowls were in males or in females? The bacillus of white diarrhea is carried by the females and is transmitted through the eggs.

PAUL E. STEINER: There seems to be no peculiar geographic distribution of Hodgkin's disease. Rural and urban inhabitants appear to be equally affected. Decreased sensitization does not seem to be associated with Hodgkin's disease. So far as I recall, the lesions along the intestinal tract were in both male and female chickens.

#### AN EXPLANATION FOR NORMAL DAILY VARIATIONS IN THE RATE OF SEDIMENTATION OF ERYTHROCYTES. EMIL T. HOVERSON.

Although the erythrocyte sedimentation test has been generally accepted both in Europe and America as a means of arriving at a diagnosis of the clinical activity of certain infectious diseases, its value in this connection has been doubted. Moreover, some fundamentals have been contradicted; for example, Pinner and his co-workers in tuberculosis disproved the belief that a rapid rate indicates an acute infection, and that a decreased rate connotes improvement. Variations in the rate have been noted by many, but since the specimens of blood were from patients who were known to be infected, the variations were ascribed to the acuteness of the disease. Pinner and Greisheimer found marked variations in normal persons.

The divergence of the opinions and reports on the subject is evidently not due to the difference in the methods used in making the tests, for Greisheimer observed a fairly close correlation between the methods of Cutler, Westergren and Linzenheimer and the procedures in each are standardized.

In an effort to arrive at definite conclusions regarding the cause of the variations in the rate, the work discussed here was undertaken. As a basis, the changes in chemistry, physiology, etc., observed by Petersen in connection with his meteorological studies might account for an actual state of the blood, and this in turn would affect the rate. Such changes which occur in normal persons have been worked out in detail, and were found to coincide with tropical or polar fronts (a polar front exists when the barometer is high and the temperature low, and a tropical front exists when the barometer is low and the temperature and the humidity high). If the chemistry, activity and other conditions change, there ought also to be a change in the rate of sedimentation of the erythrocytes. Accordingly, this work was planned to determine the variations and to correlate them with the meteorological conditions present at the time the determinations were made. The Linzenheimer method was used. Uniform conditions prevailed. All but two patients had the usual findings in dementia paralytica. One of these had a positive Wassermann reaction only of the serum; the other, a negative reaction of the serum and of the spinal fluid. The study extended from January 28 to March 1, 1934, and was so arranged that two or more patients were under observation throughout the interval. During this period nine distinct disturbances in meteorological conditions occurred. Five of these were designated as polar fronts and the

others as tropical fronts. Examination of the percentage variations in each patient's record shows that usually definite changes occurred with the weather changes. The effect of the weather changes was cumulative in two.

In certain patients the rate of settlement whether fast or slow was determined apparently by the character of the weather disturbance. Except for two patients in whom the effect was cumulative, the percentage variations in all of the patients tended to group themselves about an average percentage.

#### INVOLVEMENT OF THE MYOCARDIUM IN TUBERCULOSIS. HENRY HORN.

Although tuberculosis of the pericardium with extension to the myocardium is common, isolated myocardial tuberculosis is rare. Norris found only 6 cases in 1,764 persons with tuberculosis. Valentine, among 3,203 cases of generalized tuberculosis reported by various authors, found 7 with myocardial lesions. Only about 200 instances are mentioned in the literature. The types of myocardial involvement are: nodular, miliary and diffusely infiltrative. The nodular type is the most common. The nodules vary from pea-size to egg-size and some are so large as to obstruct the auricular flow of the blood. The number varies; in one reported case 18 nodules were found. They are usually round, yellowish-white and firm, and frequently well circumscribed. Ulceration is rare, but has been reported, and twice has been the cause of generalized miliary spread.

The miliary type is less common. Reference is made only to instances in which miliary tubercles were found exclusively in the myocardium. Up to 1906 only 41 reports of this finding were known. Since then 7 additional ones have been recorded. Much controversy has been waged over the incidence of miliary tubercles of the myocardium and generalized miliary tuberculosis. Weigert, Kaufmann and Maresch thought that the presence of miliary tubercles of the myocardium with generalized miliary tuberculosis was frequent. Such a conclusion is rendered doubtful, however, by a statistical review. Simmonds, for example, in 1879, in a study of 125 bodies with miliary tuberculosis, did not find a heart with miliary tubercles. Boltz in 1890 found 11 in 176, and Reichenbach, in 1896, found 4 in 85 bodies with tuberculosis. The tubercles either in the auricle or in the ventricle are of millet-seed size and semitransparent, and frequently follow blood vessels.

The diffusely infiltrative type is even less common and usually is associated with pericarditis. Grossly the myocardium is a white or uniformly gray to yellowish-gray firm tissue. The histologic structure occasionally presents diagnostic difficulties. Usually there is a granulation tissue with lymphocytes, polymorphonuclear leukocytes, giant cells of the Langhans type, and endothelial cells.

Some investigators, particularly the French, recognize still another form of myocardial tuberculosis: interstitial tuberculous myocarditis. The etiology is doubtful, and there are no morphologic characteristics of tuberculosis. In such cases, most authors consider the diagnosis of tuberculous myocarditis on morphologic evidence alone extremely hazardous and, therefore, this form is not considered in my classification.

Among 34 cases of generalized miliary tuberculosis in which the heart was examined carefully there were 3 with miliary tubercles of the myocardium. All three patients were within the first decade of life.

A boy, aged 7 months, had an old tuberculous bronchopneumonia and an acute miliary tuberculosis of the lungs, liver, spleen and leptomeninges. The heart was grossly normal. A granuloma histologically tuberculous was found. Acid-fast bacilli were not demonstrated by suitable stains.

A boy, aged 3 months, had tuberculous bronchopneumonia of the left lower lobe and acute miliary tuberculosis of the lungs, liver, spleen, kidneys, meninges and myocardium. The myocardium of the left ventricle had a few pearl-gray nodules the size of millet seeds. Histologically the nodules were tuberculous. A few acid-fast bacilli were found.

A girl, aged 10 years, had an old tuberculous bronchopneumonia of the right lower lobe and acute miliary conglomerate tuberculosis of the lungs, liver, spleen,

kidneys, brain and myocardium. Grossly there was nothing remarkable in the heart except a polypoid tissue in the wall of the left auricle at the mouth of the pulmonary vein. This was firm and covered with intact endocardium, but consisted of a central core of cheesy material. Microscopically the myocardium had several tissue masses, histologically tuberculous. Acid-fast bacilli were not found in sections or by concentration methods.

Most instances of miliary tuberculosis of the myocardium were in subjects of the first decade of life. This may be due to the relatively larger tissue mass examined in the smaller hearts. Perhaps, if more microscopic preparations were made of the adult myocardium, the comparative rarity of miliary tuberculosis in adults would be less. Of course, the presence of miliary tubercles in young persons may be coincidental. There is another possibility: The hearts of youths with rheumatic fever and with subacute bacterial endocarditis frequently contain Aschoff nodules. This and the demonstration of tubercles in 9 per cent of the hearts of children in this series who died of generalized miliary tuberculosis may suggest that the heart in youth is more often the seat of the granulomatous lesions of rheumatic fever and tuberculosis. This, at present, cannot be linked with an immunologic phase peculiar or common to these diseases.

#### DISCUSSION

C. A. DELANEY: Did you attempt to demonstrate tuberculosis in the skeletal muscles of these patients with tuberculous myocarditis?

P. R. CANNON: I have been interested in the localization of bacteria in various organs following intravenous injection and have noted that most of the bacteria are removed by the liver, spleen and bone marrow in thirty minutes. The best explanation seems to be that the bacteria are not removed by other tissues because the endothelial cells in them are small, have little cytoplasm, and hence are not actively phagocytic.

L. E. DAY: Tuberculosis of skeletal muscle occurs rarely in cattle. In some of these muscle infections, the viscera, in the usual gross examination, have no tuberculosis. When the infection is deep in the muscles, the fascia is involved. Tuberculous myocarditis without lesions in other tissues has been noted in cattle.

HENRY HORN: Tuberculosis of the skeletal muscle seems to be less frequent than tuberculosis of the myocardium. The skeletal muscles in these patients were not examined.

## Book Reviews

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**Parasitism and Disease.** By Theobald Smith, Director Emeritus of the Department of Animal Pathology, Rockefeller Institute for Medical Research. A publication of the Louis Clark Vanuxem Foundation. Price, \$2. Pp. 196. Princeton, N. J.: Princeton University Press, 1934.

Of recent and contemporary investigators of parasitism and infection, probably none has combined successful investigation with broad biologic interpretation in the same degree as the author of this book. This fortunate combination has established a leadership of wide influence. The book is the outcome of a desire on the part of the author "to bring together all alien invaders and parasites of the animal body and deal with them and the disturbance they produce under some unifying principle." That principle is the relation between parasitism and disease in its broadest manifestations, as determined in the main by the patient, comparative method of the naturalist. The struggle of living things to survive has established predacity and parasitism. Biologically, parasitism is a normal process, with pathologic manifestations in certain stages. In general, the relations between parasite and host are offensive and defensive, each parasite-host relation being more or less peculiar. This is the general theory that guides the presentation. In the large view, parasitism presents four phases, each with its own machinery: adjustment and adaptation to invasion, to multiplication, to emigration and to transfer to new hosts. Every successful parasite has solved its demands "in accordance with its capacities which it received from its free-living ancestors as matched against the resisting powers of the host." It is aberrant and incomplete parasitism that has greatest interest in the study of disease. "Aberrant movements are, on the whole, the most important factors in parasitism and in the diseases due to it. Aberrancy is the adventurous element in the life of the parasite, which leads either to death or to new conquests. Establishment in new hosts paves the way for the formation of new races and varieties, for the new host modifies and molds the invader until an equilibrium has been established. Aberrancy of parasites is furthermore responsible for much disease whenever they reach a more yielding host. This is almost the rule among protozoan forms. Most epidemics or pandemics are probably due to strayed parasites. A study of epizootics among small experimental animals kept together in large numbers tends to show that such diseases are most virulent at the start and gradually change their characters, becoming less infectious, more chronic, non-fatal and topographically altered as regards the incidence of lesions. The epidemic is probably the first sign of a straying of parasites from either near or more distantly related hosts or from an immune group to a susceptible group of the same species or race."

Medical science has been most concerned with the multiplication of the parasite within the host, that is, in the immediate host-parasite conflict. The attack on this stage in the parasitic cycle in order to save life and prevent infection has resulted in the great modern advances in the prevention and the treatment of infectious diseases. In the evolution of parasitism, offensive and defensive weapons develop on the part of both parasite and host. In the case of the host, this development is illustrated by the formation of specific antibodies and the development of phagocytic cells.

The last four chapters of this remarkable book deal with the variation and mutation among parasites, the survival of parasites and movement from host to host, epidemiology and the utilization of discoveries in parasitism. In these and in the other chapters the complex relations of parasitism to disease are discussed with great clearness and deep insight from a broad, biologic point of view. All forms of parasites receive attention—worms, protozoa, bacteria and invisible viruses. It is a stimulating book, which will interest a much wider circle than those interested merely in the medical or technical aspects of the subject. Two

more quotations may be permitted: "The final suppression [of epidemics] presupposes a world organization of human society without wars, and disarmed, such as the most pronounced idealist of today can scarcely conceive, but towards which human society must tend to survive in the struggle with animal and plant life, microscopic and ultramicroscopic" (page 168). . . . "The objectives of research as a mere accumulation of data or the display and parading of acquired knowledge in a world otherwise in motion is outmoded. Penetration into mysteries may thrill the penetrator and occasionally others, yet this is not enough. Discoveries and inventions must be made to yield some contribution towards that rather vague goal, the welfare of mankind. More specifically the study of parasitism and disease appeals to the human desire to live and be free from disease and it is this strong urge that furnishes ample stimulus to the altruist as well as to the charlatan. Parasitism is greatly favored by large numbers of individuals insofar as they offer the best opportunities for new parasites through trial and error methods to arise or for existing ones to develop a more complete adaptation. Large numbers of hosts furnish the starting points of epidemics and nature reduces the superabundance of wild life through disease. Nature may be said to abhor a crowd" (page 169).

**Les ultraviruses pathogènes saprophytes: techniques d'études, caractères physiques et biologiques, maladies à ultraviruses, clinique, anatomic pathologique, épidémiologie, immunité.** By Paul Hauduroy, Préparateur à la Faculté de Médecine de Paris. Price, 60 francs. Pp. 462. Paris: Masson & Cie.

An earlier book by the author on filtrable viruses ("Les ultraviruses et les formes filtrantes des microbes") was published in 1929. Since then knowledge of so-called filtrable or ultramicroscopic viruses—the ultraviruses—has grown with amazing rapidity. In this book Hauduroy presents a comprehensive review of the present state of knowledge in this field. The first part of the book deals with the technic of filtration, ultrafiltration, cataphoresis, adsorption, cultural methods and staining. The second part is devoted to a description of the ultraviruses—saprophytic and pathogenic, of plants, bacteria, insects, birds and mammals. In the third and last part the principal characteristics of the ultraviruses are reviewed. Under the virus diseases in man are grouped smallpox and amas, epidemic poliomyelitis, herpes, encephalitis, chorea, yellow fever, dengue, inguinal lymphogranulomatosis (inguinal granuloma is not discussed), influenza, coryza, warts, molluscum contagiosum, mumps, trachoma and measles. Rabies and vaccinia are described as primary in animals. In the section on sheep pox (*claveléc*) is an interesting citation from Pourquier (*Compt. rend. Acad. d. sc.* **101**:863, 1885; *ibid.* **104**:703, 1887), in which he described independently allergic phenomena in this disease that correspond exactly to the allergic phenomena described by Jenner, Boyce and others in smallpox and vaccinia more than one hundred years ago and by Pirquet in 1906. Pourquier's description is as follows: "If, during six successive days the same subject is inoculated with the same virus, the first four punctures only reach the stage of pustules, and in all development is complete and secretion is greatest from the fourteenth to the fifteenth day from the beginning of the experiment. There is consequently an inequality in the duration of the evolution, the first puncture showing secretion within fifteen days, the second at fourteen days, the third at thirteen days and the fourth at twelve days. This inequality in the duration of the evolution modifies the dimensions of the pustules, the diameters of which decrease. It appears from these observations that after the first twenty-four hours the first inoculation had already modified the soil (*terrain*). This influence is accentuated the following days, and immunity is acquired from the sixth to the seventh days: On the sixth day there is at least a papule, on the following days nothing. These differences in the evolution are due to a modification of the soil at the beginning of immunity."

Hauduroy's book gives an instructive and orderly account of what is now known about the ultraviruses. It will be of value to all who are interested in this new field of investigation.

## Books Received

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A PRACTICAL TREATISE ON DISEASES OF THE SKIN FOR THE USE OF STUDENTS AND PRACTITIONERS. Oliver S. Ormsby, M.D., Clinical Professor and Chairman of the Department of Dermatology, Rush Medical College of the University of Chicago. With Revision of the Histopathology in this Edition by Clark Wylie Finnerud, B.S., M.D., Assistant Clinical Professor of Dermatology, Rush Medical College of the University of Chicago. Fourth edition. Cloth. Price, \$11.50. Pp. 1288, with 622 illustrations. Philadelphia: Lea & Febiger, 1934.

ACTIVE IMMUNIZATION AGAINST DIPHTHERIA: ITS EFFECT ON THE DISTRIBUTION OF ANTITOXIC IMMUNITY AND CASE AND CARRIER INFECTION. Sheldon F. Dudley, Percival M. May and Joseph A. O'Flynn with a note by J. Orr Ewing. Medical Research Council, Special Report Series, No. 195. Price, 3 s. Pp. 140. London: His Majesty's Stationery Office, 1934.

RECENT ADVANCES IN PATHOLOGY. Geoffrey Hadfield, M.D., F.R.C.P., Professor of Pathology in the University of Bristol, and Lawrence P. Garrod, M.A., M.D., B.Ch., Bacteriologist and Lecturer in Bacteriology, St. Bartholomew's Hospital. Second edition. Cloth. Price, \$4. Pp. 457, with 69 illustrations. Philadelphia: P. Blakiston's Son & Company, Inc., 1934.

PATHOLOGIE UND KLINIK DER GRANULOSAZELLTUMOREN. Dr. Walter Schiller, Assistent der II. Universitäts-Frauenklinik (Prof. Dr. Wilh. Weibel) in Wien. Price, 16 marks. Pp. 197, with 136 illustrations. Vienna: Wilhelm Maudrich, 1934.

CLINICAL MISCELLANY. The Imogene Bassett Hospital, Cooperstown. Contributors: Floyd J. Atwell, M.D.; C. Lee Buxton, M.D.; Francis F. Harrison, M.D.; Monroe A. McIver, M.D.; Charles C. McCoy, M.D.; G. M. Mackenzie, M.D.; Marjorie F. Murray, M.D.; Robert M. Pike, Ph.D.; John H. Powers, M.D.; T. J. Abernethy, M.D. Price, \$3. Pp. 206, with 37 illustrations. Springfield, Ill.: Charles C. Thomas, Publisher, 1934.

REPORT OF ACTIVITIES FOR PERIOD ENDING DECEMBER 31, 1933. The International Cancer Research Foundation, 1616 Walnut Street, Philadelphia. Chartered in Pennsylvania, April 25, 1932. Organized in Philadelphia, June 8, 1932. Pp. 43.

# ARCHIVES OF PATHOLOGY

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## PATHOLOGY OF THE CENTRAL NERVOUS SYSTEM IN CANINE BLACK TONGUE

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The etiology of pellagra, which was apparently established by Goldberger on the basis of a vitamin deficiency, has become once more a matter of controversy. This is largely due to the unsatisfactory results of therapy, for not infrequently the oral use of materials rich in vitamin B complex and hence presumably containing the pellagra-preventing factor described by Goldberger and his co-workers fails to cure. The work of that group was based, to a large degree, on studies of an experimentally induced condition in dogs known as black tongue. This canine disease is marked by stomatitis, glossitis, salivation and diarrhea, all these symptoms being prominent also in pellagra. According to Wheeler, black tongue and pellagra are the same disease, "on account of their seasonal and geographical incidence, their common cause and similar course, their identical pathological changes and their equal response to the same therapeutic and preventive measures." Analysis of the published studies, however, shows that in one pathologic respect the two conditions cannot be stated to be identical. The discrepancy is in the presence of pathologic alterations of the central nervous system in pellagra and their absence in black tongue. This discrepancy is the more important because of recent studies showing the effect of lack of the vitamin B complex on the production of lesions of the central nervous system marked by loss of myelin. If, as is claimed by the Goldberger school, pellagra is due to the absence of the thermostable fraction of the vitamin B complex and if such a lack is causative in the production of demyelinating lesions of the central nervous system, both black tongue and pellagra should be associated with pathologic alterations of the brain and spinal cord. A large number of histologic studies have given incontrovertible proof that degenerative lesions both of myelin and of nerve cells are present in pellagra. The prominence in pellagra of symptoms referable to nerve lesions is further evidence of involvement of the

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central nervous system. It is difficult to reconcile the lack of parallelism in the neuropathologic studies of black tongue and pellagra in view of the remarkable pathologic and symptomatic similarity in other respects.

Since the studies of the neuropathology of canine black tongue are not extensive or detailed, changes similar to those found in the central nervous systems of persons dying of pellagra may have been overlooked. Accordingly, the brains and spinal cords of twelve dogs dying of acute black tongue were studied by modern neuropathologic methods. The changes observed were similar in many respects to those seen in pellagra. These changes were also similar to those described as occurring in animals kept on diets deficient in the vitamin B complex. The presence of degenerative lesions of the central nervous system in pellagra may explain, to a certain extent, the difficulties attending therapy in pellagra. These pathologic alterations also make up a further link in the chain of evidence associating deficiency in a factor closely allied to the vitamin B complex with the etiologic agent of pellagra, black tongue and certain conditions in man marked by loss of myelin in the central nervous system.

#### REVIEW OF THE LITERATURE

The only comprehensive and detailed studies of the pathologic changes in acute black tongue are those of Denton.<sup>1a</sup> The animals were killed and examined immediately. The nervous systems were studied by general pathologic methods used as a routine. Aside from alterations of the nerve cells, no significant observations were reported.

The literature on pellagra contains a number of references to alterations of the central nervous system. Denton<sup>1b</sup> observed only indefinite changes in the nerve cells in the more acute cases. Singer and Pollock<sup>2</sup> described chromatolysis of nerve cells, astrocytosis and satellitosis, as well as destruction of nerve fibers. Langworthy<sup>3</sup> observed diffuse loss of nerve fibers and changes in the nerve cell. Castellani and Chalmers<sup>4</sup> observed degeneration of nerve cells and disappearance of fibrils in the posterior and lateral columns. Singer<sup>5</sup> observed diffuse degeneration of fibers in the white matter and pronounced changes in the nerve cells of the cerebrum and cord. Sclerosis was observed in the more chronic cases. Klauder and Winkelman<sup>6</sup> described so-called "central neuritis"

1. Denton, J.: (a) Am. J. Path. **4**:341, 1928; (b) Am. J. Trop. Med. **5**:173, 1925.

2. Singer, H. D., and Pollock, L. J.: Arch. Int. Med. **11**:565, 1913.

3. Langworthy, O. R.: Brain **54**:291, 1931.

4. Castellani, A., and Chalmers, A. J.: Manual of Tropical Medicine, ed. 3, London, Baillière, Tindall & Cox, 1919.

5. Singer, H. D.: Arch. Int. Med. **15**:121, 1915.

6. Klauder, J. V., and Winkelman, N. W.: J. A. M. A. **90**:364, 1928.

in the brains of twelve patients with pellagra. The pathologic changes included swelling of the cells, with disappearance of Nissl bodies, and an increased lipoid content throughout the entire central nervous system. Wilson<sup>7</sup> observed chromatolysis of the nerve cells of Clarke's column and the anterior horns, as well as degeneration of the white matter. Vedder<sup>8</sup> described extensive myelin degeneration of the cord, with chromatolysis and pigmentation of nerve cells. Kozowsky<sup>9</sup> and Winkelmann<sup>10</sup> observed a deposit of fat in the nerve cells in pellagra, as did Sandwith<sup>11</sup> and Pentschew.<sup>12</sup> Reed and Ash<sup>13</sup> described column degeneration of the spinal cord in sprue, a condition having certain symptomatic similarities to pellagra. The etiologic relationship of deficiencies in the water-soluble vitamin to pathologic alterations in the central nervous system has been observed since the early studies of beriberi.<sup>14</sup> More recent investigation employing mammals has brought forward additional evidence. Gildea, Kattwinkel and Castle<sup>15</sup> made histologic studies of the central nervous systems of dogs maintained on a synthetic diet deficient in the vitamin B complex. Extensive degenerative changes in the myelin sheaths were observed. Faulty staining technic made their results difficult to interpret. It does not appear, however, that lesions followed deprivation of any single component of the vitamin B complex. Zimmerman and Burack<sup>16</sup> described lesions of myelin in animals maintained on diets deficient in the thermolabile, and probably also in the thermostable, fractions of the vitamin B complex. The evidence is clear that lack of the water-soluble vitamin gives rise to demyelinating lesions of the central nervous system, but the evidence for the rôle of any particular component of the vitamin B complex is as yet unsatisfactory.

#### METHODS

Dogs of good size and of mongrel breed, largely short-haired, were employed. The animals were kept under uniform conditions in individual cages with bedding

7. Wilson, S. A. K.: Proc. Roy. Soc. Med. (Neurol. Sect.) **7**:31, 1914.
8. Vedder, E. B.: Arch. Int. Med. **18**:137, 1916.
9. Kozowsky, A. D.: Arch. f. Psychiat. **49**:204, 556 and 873, 1911-1912.
10. Winkelmann, N. W.: Ztschr. f. d. ges. Neurol. u. Psychiat. **102**:38, 1926.
11. Sandwith, F. M.: J. Path. & Bact. **7**:460, 1901.
12. Pentschew, A.: Ztschr. f. d. ges. Neurol. u. Psychiat. **118**:17, 1928.
13. Reed, A. C., and Ash, J. E.: Arch. Int. Med. **40**:787, 1927.
14. (a) Eijkman, C.: Virchows Arch. f. path. Anat. **148**:523, 1897; (b) Vedder, E. B., and Clark, E.: Philippine J. Sc. **7**:423, 1912; (c) Findlay, G. M.: J. Path. & Bact. **24**:175, 1921.
15. Gildea, E. F.; Kattwinkel, E. E., and Castle, W. B.: New England J. Med. **202**:523, 1930.
16. Zimmerman, H. M., and Burack, E.: Arch. Path. **13**:207, 1932.

of shavings. No particular care was taken to avoid coprophagy. The diet was composed of the following ingredients:

White corn-meal.....	6,000 Gm.
California black-eyed peas.....	750 Gm.
Casein .....	900 Gm.
Cod liver oil.....	450 cc.
Cottonseed oil.....	450 cc.
Rice polishings.....	600 Gm.
Calcium carbonate.....	450 Gm.
Sodium chloride.....	150 Gm.

The corn-meal, peas and casein were mixed and cooked for two hours in a steam cooker. The remaining ingredients were then added and thoroughly mixed. The dogs were fed six days a week and were allowed to eat as much as they chose. They were weighed at weekly intervals.

Treatment was instituted only when the symptoms were so acute and severe that it seemed doubtful whether the animal would survive in case no therapy was given. In treatment, 200 Gm. of raw beef was fed daily if the animal would eat. If not, a rice polishings concentrate prepared according to the method described by Evans and Lepovsky<sup>17</sup> was administered by stomach tube. Occasionally a brewers' yeast concentrate was given. Successful therapeutic results were so difficult to obtain when the disease was at its height that it was often impossible to confine the treatment to some simple material.

In general, the diet was well taken. Control animals which were given 4 Gm. daily of liver extract-Lilly ate no more than the dogs maintained on the unsupplemented diet and remained in perfect health.

Animals which ate sparingly of the special diet also failed to acquire the disease under investigation, but frequently died of intercurrent infections. It seemed clear that whatever symptoms developed were due to the absence of an accessory food factor in the diet and not to lack of salts, metallic elements, particular proteins or caloric intake.

*Staining Technic.*—Autopsies were done on seven of the animals within one-half hour after death. In the other five the period post mortem was not known exactly, since the bodies were found in the morning. The brains and spinal cords were fixed at autopsy in a diluted solution of formaldehyde, U.S.P. (1:10). From three to five months after the death of the dogs the material was sent to the Neurological Laboratory of the Boston City Hospital. Blocks were cut at once and put into Müller's solution<sup>17a</sup> for staining by the Weigert method. After six weeks in Müller's solution the blocks were embedded in pyroxylin (celloidin) and cut and stained by the conventional Weigert-Pal technic. Other blocks were washed and passed through alcohol and pyroxylin. Sections from these blocks were stained with toluidine blue. Stains for fat were made on frozen sections by the Herxheimer method, using scarlet red. For the Alzheimer-Mann stains, blocks were cut and mordanted in "Gliabeize."<sup>17b</sup> Mann's solution (equal parts

17. Evans, H. M., and Lepovsky, J.: *J. Nutrition* 3:353, 1931.

17a. Müller's solution is: potassium bichromate, 2 Gm.; sodium sulphate, 1 Gm., and water, 100 cc.

17b. The formula for Gliabeize is as follows: potassium bichromate, 5 Gm.; fluorochrome, 2 Gm., and water, 100 cc. (Mallory, F. B., and Wright, J. H.: *Pathological Technique*, ed. 8, Philadelphia, W. B. Saunders Company, 1924, p. 155).

of eosin and methyl blue) was used for staining the frozen sections. This method stains normal axis-cylinders a clear blue, in striking contrast to the red myelin and stroma.

Examination of the sections stained by the Weigert method disclosed widespread changes; hence, other stains were employed in an attempt to confirm the

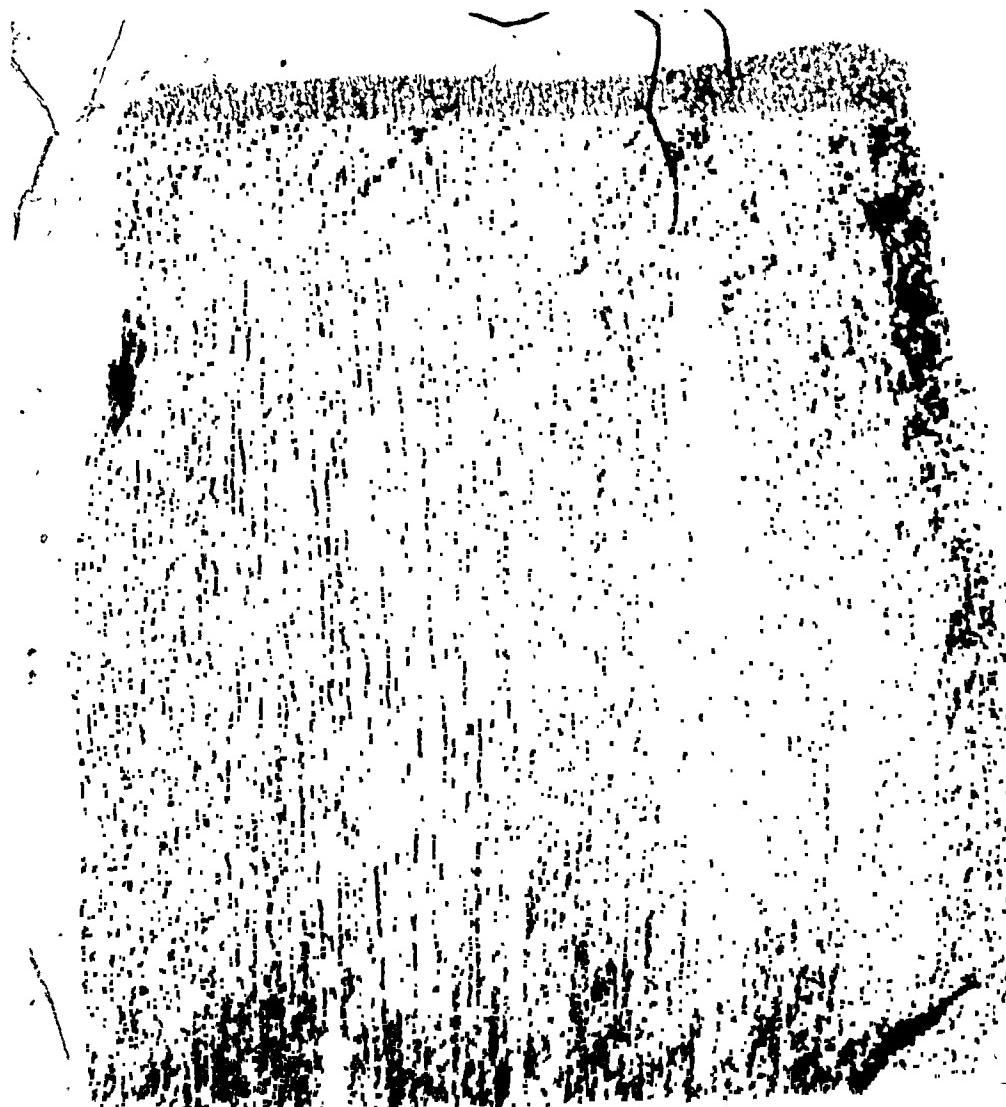


Fig. 1 (dog 9).—Photomicrograph of a longitudinal section from the spinal cord of a dog which lived for two months on the black tongue-producing diet and died showing stomatitis, glossitis, salivation, diarrhea, tremor and ataxia. The cord was fixed within one hour post mortem. The lesion is widespread and involves diffuse and irregular degeneration of myelin; much débris is present, especially along the margins of the section. Weigert-Pal stain; low power magnification ( $\times 10$ ).

observations. Some of the cords were cut in frozen sections and stained by the Spielmeyer method. These sections showed exactly the same condition of wide-

spread degeneration that appeared in the material stained by the Weigert technic. The cords stained by both the Weigert and the Spielmeyer method showed the lesions to be generally disposed around the margins of the sections, with the more central areas showing relatively little alteration. There was no perceptible differ-



Fig. 2 (dog 4).—A longitudinal section from the spinal cord of a dog which lived for three and a half months on the black tongue-producing diet. At death the dog showed glossitis, stomatitis, diarrhea and tremor. The cord was fixed within one hour post mortem. The section shows widespread degeneration of the myelin, with few normal sheaths; there is much débris at the right of the section. Weigert-Pal stain; high power magnification ( $\times 185$ ).

ence between the sections from dogs the cords of which were fixed at once and those which may have been dead several hours before the autopsy.

Because the stains for fat showed no changes in the white matter, other attempts were made to determine the nature of the products of degeneration. Nile blue sulphate was used to stain any free fatty acids present, but none was demon-



Fig. 3 (dog 5).—Photomicrograph of cells in the anterior horn of a dog which lived three months on the black tongue-producing diet and died showing stomatitis, glossitis, salivation, diarrhea and tremor. The cord was fixed within one hour post mortem. The anterior horn cells show condensation of the Nissl substance and encrustations. Cresyl violet stain; immersion in oil;  $\times 380$ .

strated. It was considered impossible to use Marchi's method because of the type of fixation employed. The technic described by Donaggio was used instead. This

method is stated to demonstrate very early myelin degeneration, staining the pathologic sheaths black in a colorless field. The picture thus produced is similar to that made by the Marchi method, but is said to give a positive stain of

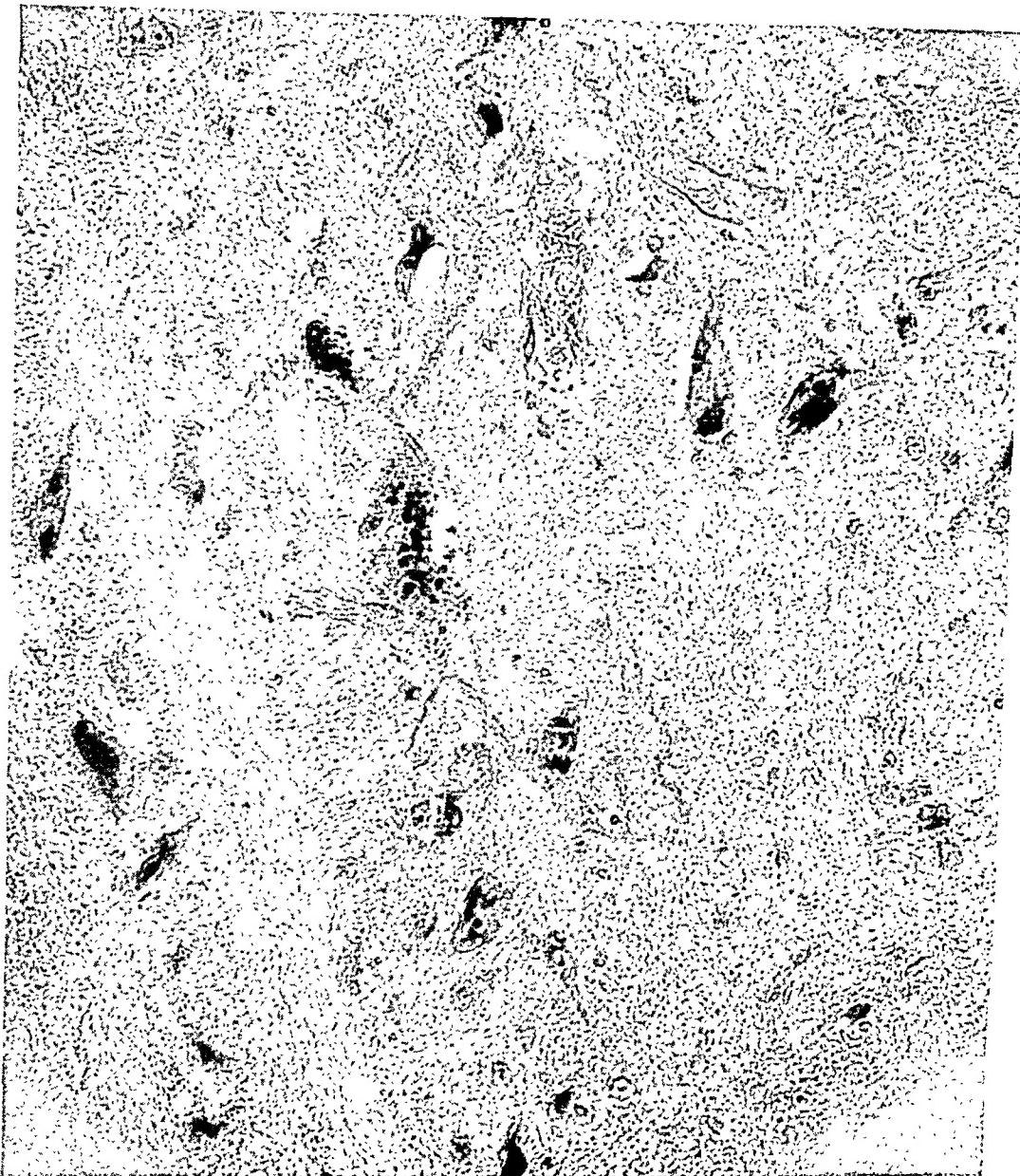


Fig. 4 (dog 3).—Nerve cells from a longitudinal section of the cord of a dog which lived for three and a half months on the black tongue diet and showed stomatitis, glossitis, salivation and diarrhea at death. The cord was fixed within one hour post mortem. The section shows atrophy and degeneration of the nerve cells; two cells in the left center show condensation of the Nissl substance and encrustations. Cresyl violet stain; immersion in oil;  $\times 380$ .

degeneration within from three to five days. The material embedded in pyroxylin, which was prepared for staining by the Weigert method, was suitable for use



Fig. 5.—*A*, anterior horn cells from the cord of one of the dogs with black tongue, showing the large deposits of lipoid pigment, which stained green, with chromatolytic and lipoid changes in the nuclei; cresyl violet stain; reduced from  $\times 450$ . *B*, anterior horn cells from one of the dogs with black tongue, showing the lipoid deposits, which stained red; Herxheimer scarlet red stain; reduced from  $\times 450$ . *C*, longitudinal section of the cord of a normal dog, showing continuous axis-cylinders, which stained blue, and regularly arranged stroma; Alzheimer-Mann stain; reduced from  $\times 450$ . *D*, longitudinal section from the cord of one of the dogs with black tongue, showing broken, slightly tortuous axis-cylinders, which appeared reddish on staining, and pale, edematous, irregular stroma; Alzheimer-Mann stain; reduced from  $\times 450$ .

(having been mordanted in Müller's solution before embedding), and cross-sections were cut and stained with hematoxylin as for the Weigert method. The sections were immersed in neutral copper acetate for approximately thirty minutes and then differentiated with oxalic acid and potassium permanganate in the conventional Pal manner. In the description of this technic by Herxheimer<sup>18</sup> it is stated that the normal sheaths are uncolored, except on the extreme margins of the sections. In the controls observed in this study it was found that a narrow rim of black sheaths occurred along the margins of all sections, and this rim was somewhat variable in thickness in different sections. In the animals dying of black tongue this rim of black sheaths was invariably wider than in the controls and coincided well with the apparent location of the lesion as determined by the Weigert and Spielmeyer stains. Since the stain is relatively unknown and since it was impossible to examine a large series of spinal cords from normal animals, it is considered inadvisable to lay too much emphasis on the results obtained. They are reported here simply because they checked in general with the other observations.

Dog 4.—The diet was begun on Jan. 7, 1932. On February 24 the chronic, diffuse, granular reddening of the entire buccal and lingual mucous membranes was noted. Complete achlorhydria and chronic diarrhea developed. On March 1 there was a severe attack of acute stomatitis and glossitis, which persisted for four days and from which the animal recovered. On April 1 diffuse, chronic glossitis and stomatitis set in, which progressed steadily, with diarrhea. On April 10 tremors and weakness developed. Rice polishings vitamin B<sub>1</sub> concentrate (Block) was administered intravenously. The symptoms continued and progressed until April 18, when the animal was comatose. On April 19 it was found dead. Autopsy was performed immediately.

*Weigert Stain*.—Cross-sections showed the periphery of the cord to be less dense than the center. The tissue along the periphery was ragged and contained many clumps of degenerated myelin. The longitudinal sections appeared irregularly stained. Many clumps of myelin and much débris were present. The cord appeared to be definitely diseased, since almost no part of the sections had a normal appearance.

*Herxheimer Stain*.—The cross-sections showed a rare droplet of fat in the white matter. The amount was so small as not to be significant, and it probably was not in phagocytic cells. The nerve cells contained a considerable amount of fat. The longitudinal sections showed similar changes.

*Toluidine Blue Stain*.—In the cross-sections the meninges appeared normal. The white matter was normal, except for glia cells with hyperstaining granular cytoplasm. No infiltration or gliosis was apparent. In the gray matter the nerve cells were faded, vacuolated and degenerating. Many of them were surrounded by satellites, but, on the whole, satellitosis was not marked. The longitudinal sections differed in no particular from the cross-sections.

*Alzheimer-Mann Stain*.—Cross-sections showed the periphery to be much less dense than the center. There was no variation in staining of the axis-cylinders and no infiltration. The longitudinal sections were irregularly stained, particularly at the periphery. There was considerable variation in staining of the axis-cylinders from blue through brilliant red. Some infiltration was present, and phagocytes in small numbers were present in the tissue.

18. Herxheimer, G.: Technik der pathologisch-histologischen Untersuchung, Wiesbaden, J. F. Bergmann, 1912, p. 321.

*Donaggio Stain.*—The sections were typically positive. There were aggregations of black spots around the periphery, more than were seen in the controls.

*Summary.*—Examination of the stained sections showed: results with Weigert and Alzheimer-Mann methods, +++; poliomyelopathy, ++.

Dog 9.—The diet was begun on Feb. 26, 1932. On April 4 a red, inflamed patch of mucous membrane appeared under the upper lip. This extended and became progressively worse until within a few days there was a mild, chronic injection of the entire lingual and buccal mucous membranes. Diarrhea set in and persisted until death. On April 19 severe stomatitis and glossitis were noted, with salivation, tremor and ataxia. On April 24 the animal was found dead. Autopsy was performed immediately.

*Weigert Stain.*—The cross-sections showed fading and loss of myelin, but degenerated myelin was not present. Longitudinal sections showed a striking lesion of myelin. There was complete loss of staining in the involved area, with heading, swelling of sheaths and discontinuity. Much débris was present.

*Herxheimer Stain.*—Cross-sections showed no fat. Longitudinal sections showed no fat in the white matter but small amounts in the nerve cells and glia cells of the gray matter.

*Toluidine Blue Stain.*—Cross-sections showed the meninges to be normal. The glia cytoplasm was deeply stained. The myelin and axis-cylinders had taken a pale blue stain. In the gray matter the nerve cells were undergoing every form of degeneration. Some were vacuolated, with the nuclei gone. Some were dark-staining or vitreous. The longitudinal sections were not different from the controls, except for some satellitosis.

*Alzheimer-Mann Stain.*—The cross-sections showed many unstained areas, especially around the periphery, but no alteration in the staining of the axis-cylinders was present. Longitudinal sections showed an unquestionable lesion of myelin. Little normal myelin remained. The axis-cylinders were fragmented. There was no infiltration.

*Donaggio Stain.*—No convincing change was observed.

*Summary.*—Examination of the stained sections revealed: results with the Weigert method, +++, and with the Alzheimer-Mann method, ++ poliomyelopathy, +.

Dog 12.—The diet was begun on March 9, 1932. Inability to secrete free hydrochloric acid developed. On April 3 stomatitis, glossitis and salivation set in, which continued with remissions and exacerbations for ten days. On April 17 tremor, ataxia, and weakness appeared; these became progressively worse. On April 21 the animal was found dead. Autopsy was performed immediately.

*Weigert Stain.*—Cross-sections showed no significant changes. The longitudinal sections showed striking changes; most of the myelin sheaths were intact, but there were areas of irregular staining, with clumps of degenerated myelin.

*Herxheimer Stain.*—Cross-sections showed a few droplets of fat in the perivascular spaces of the white matter. There was much fat in the nerve cells and neuroglia of the gray matter, and some in the perivascular spaces. Longitudinal sections showed droplets of fat in most of the perivascular spaces in the gray matter and in almost all the nerve cells. Fat was present in some neuroglia cells as well.

*Toluidine Blue Stain.*—Cross-sections showed the meninges to be normal. The white matter was normal, except for neuroglia cells with hyperstaining cytoplasm

and stained processes. The nerve cells were vacuolated and disintegrating; many contained greenish pigment. The Nissl bodies were small and dustlike. Satellitosis was not marked.

*Alzheimer-Mann Stain.*—Cross-sections showed a variation in the intensity of staining. The meninges were slightly thickened and engorged. The white matter showed some irregularity of staining, especially at the periphery. The longitudinal sections were not very different from the controls.

*Donaggio Stain.*—Every section showed blackened spots and sheaths around the periphery, spreading inward toward the gray matter.

#### Tabular Summary of Results with Various Methods of Staining\*

Dog	1 Weigert Stain	2 Alzheimer-Mann Stain	3 Herkheimer Stain	4 Toluidine Blue Stain	5 Donaggio Stain
1	++	+	++	+	—
2	+	++	+	+	—
3	—	+	—	+	—
4	+ <sup>+</sup>	+++	++	++	+
5	++	+++	+	+	+
6	++	+++	—	+	—
7	++	++	++	—	++
8	++	+	++	+	+
9	+ <sup>+</sup>	++	+	+	—
10	+	+	++	+	—
11	++	+	+	+	—
12	+	—	+++	+	++

\* In column 1, — indicates that the results were not different from those obtained by the same method in control animals; +, that there was fading of some myelin sheaths, i.e., a definite failure to take the hematoxylin stain; ++, that there was a more advanced degree of the same change; +<sup>+</sup>, that large areas of fading were present with *Markballen* and phagocytosis.

In column 2, — indicates that the picture was comparable to that shown by the controls; +, that a definite number of axis cylinders took a red stain; ++, that a more extensive process was noted; +<sup>+</sup>, that a large proportion of the axis cylinders were red and that the stroma was largely disrupted.

In column 3, — indicates that the sections were like the controls; +, that more fat was present in some nerve cells than in any control; ++, that an intermediate lesion was observed; +<sup>+</sup>, that fat was present in at least 80 per cent of the nerve cells and that a small amount was present in phagocytes in the white matter.

In column 4, — indicates that a section was like the controls; +, that the cytoplasm of the glia cells was stained and that various forms of degeneration were present in about 25 per cent of the nerve cells (these changes consisted of the axonal reaction, swelling of the cell, chromatolysis, vacuolation, the presence of a greenish pigment and nuclear changes consisting of eccentrically placed or absent nuclei); +<sup>+</sup>, that the changes just described were present in approximately 50 per cent of the nerve cells and that some of the nerve cells showed encrustations.

In column 5, — indicates a picture falling within the wide range of the normal controls; +, means that the band of myelin sheaths around the margin of the cross section of the cord which stained black was wider than in any control; +<sup>+</sup>, that there was a wide band of black-stained marginal sheaths and also that black sheaths were present in areas approaching the central gray matter.

*Summary.*—Examination of the stained sections revealed: results with the Weigert method, +, and with the Alzheimer-Mann method, —; poliomyelopathy, ++.

#### GENERAL SUMMARY

In eleven of the twelve animals studied, lesions were demonstrated by the Weigert method of staining. The lesions were in general not striking and had apparently produced no definite reaction in either the neuroglia or the cells taking part in inflammatory processes. The cytoplasm of the neuroglia cells throughout the entire series had stained, and in some instances was quite granular. This may have been the beginning of a reaction on the part of these elements.

In all the myelin stains the structural change was, for the most part, slight and consisted of irregularity, swelling and shrinking of the fibers. Only in occasional sections was there actual breakdown into droplets, and the droplets were probably never phagocytosed (dog 6 may be an exception to this). There was no definite free fat present in the white matter of any dog, except possibly dog 12.

The Alzheimer-Mann stain was used here chiefly to determine the condition of the axis-cylinders, although it also stained the myelin and stroma. Of the twelve dogs, eleven showed alteration in the axis-cylinders, manifested by a reddish stain instead of the normal blue; the axis-cylinders that were present in the lesions were swollen, broken up and tortuous, staining from purple through brilliant red.

Poliomyelopathy, indicating a change in the nerve cells, occurred in eleven of the twelve dogs. It was characterized by various types of degeneration in the nerve cells and the presence of fat in the cells. In dog 3 poliomyelopathy was considered present, although fat was not demonstrated because the Nissl stain showed such marked changes.

The significance of the results obtained with the Donaggio method of staining is unclear. The control animals showed such a wide range of variation that sections from the dogs with black tongue had to be interpreted with great caution. Only sections showing a definite increase over the maximum change seen in the controls were reported positive.

Interpretation of data such as those presented is difficult. The changes described appear to be of an early acute type and perhaps autolytic or chemical. The most marked alterations present were of the myelin sheaths, which took the stain irregularly and showed various forms of a degenerative process. The other components of the central nervous system also showed consistent, though mild, alterations, which were in general considered to be pathologic. In brief, there was evidence of a general disturbance of the structure of the central nervous system.

The etiology of the changes described is obscure. The addition of rice polishings to the diet should have insured an adequate supply of antineuritic vitamin B<sub>1</sub>. It is known that the symptoms associated with the pathologic lesions may be prevented by the addition to the diet of a variety of food substances. Many of these substances are known to be rich in the thermostable factor (vitamin B<sub>2</sub> [G]) effective in promoting the growth of rats maintained on an otherwise adequate diet. Examples are meat, milk and yeast. A commercial yeast concentrate known to be poor in the growth-promoting factor is also effective as a prophylactic. Liver extract-Lilly is also effective in this regard. Lack of vitamin B<sub>1</sub> or B<sub>2</sub> (G) or both does not give rise to the clinical syndrome described. It appears, therefore, that the condition is due to lack of an essential food substance which is as yet unidentified but which appears to be frequently associated with vitamin B<sub>2</sub> (G).

Comparison of the histologic alterations described in the protocols with those which have been found in the central nervous systems of human beings dying of pellagra shows a striking similarity. In view of the like pathology of other organs, the similar symptoms and the almost identical effective preventive and therapeutic measures, it seems likely that canine black tongue and human pellagra may be related etiologically. It appears that the deficiency which is causative in these two conditions gives rise to fairly characteristic lesions of the central nervous system, even though symptoms referable to that system may be slight and the disease process apparently of brief duration.

#### CONCLUSIONS

1. Lesions of the central nervous system are present in animals dying of acute black tongue.
2. The lesions are characterized by disintegration of myelin, alterations of the axons and degenerative changes of nerve cells.
3. The changes resemble those described in pellagra and in animals deficient in the vitamin B complex.

# STUDIES IN ATHEROSCLEROSIS: CHEMICAL, EXPERIMENTAL AND MORPHOLOGIC

I AND II. RÔLES OF CHOLESTEROL METABOLISM, BLOOD PRESSURE AND  
STRUCTURE OF THE AORTA; THE FAT ANGLE OF THE AORTA  
(F.A.A.), AND THE INFILTRATION-EXPRESSION THEORY  
OF LIPOID DEPOSIT

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## INTRODUCTION

Concerning the pathogenesis of atherosclerosis of the aorta there have been three important theories, viz., that it is infectious, degenerative or metabolic.

The first theory was championed by Virchow who believed the process to be partially infectious, and he spoke of an "Endarteritis chronica nodosa deformans." His idea was that the infectious process so altered the wall of the vessel that imbibition of fat from the serum of the blood stream followed and fat was deposited there. His theory in modified form still has some adherents (Klotz,<sup>b</sup> Saltykow \*) but more generally has been discredited.

The infectious theory was then replaced by the degenerative one (Marchand, Jores, Thomas, Aschoff<sup>c, d, e</sup>), and here again two different stands were taken. Thoma's concept was that the media underwent a primary degeneration or weakening (*angiomalacia*) followed by compensatory proliferation of the intima (*Angiosklerose*) (see also Faber, Adami and Krauss, Beitzke, cited by E. Kaufmann). Marchand and Jores found no microscopic changes in the media of the aorta and ascribed the primary cause of atherosclerosis to a fatty degeneration of the intima. Thus, Marchand changed Lobstein's terminology of arteriosclerosis to that of atherosclerosis. The source of the fat of the intima was then shown by Aschoff not to be a degeneration of the intima but rather an infiltration from the nutrient blood plasma ushered in from the lumen of the aorta, thereby reviving Virchow's original imbibition theory (Ribbert, Stumpf, Hueck, cited by Jores).

The cause of the primary degenerative process was considered to be the outcome of the excessive functional demand made on the aorta (Thoma, Marchand, Beneke, Moschcowitz, Lange,<sup>b</sup> Rühl) or the conse-

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\* The bibliographic references will be found at the end of the fourth article, which will be published in the November issue.

From the Pathologic Institute, Freiburg, Germany; Dr. L. Aschoff, director; the Pathologic Institute of the Cook County Hospital, Chicago; Dr. R. H. Jaffé, director, and the chemical division of the Pathologic Institute, Freiburg, Germany; Dr. R. Schoenheimer, formerly director.

quence of the wear and tear of age (Romberg,<sup>b</sup> Wells). Accordingly, the entire process of atherosclerosis was set down as inevitable.

The metabolic trend of thought takes its initiative from Aschoff,<sup>a, b</sup> who showed that the doubly refractive bodies in atheromatous aortas were similar to cholesterol esters, and that they infiltrated from the blood stream; and its challenged verification, from Anitschkow and Chalatow, also Wacker and Hueck, who produced atheromatous lesions in the rabbit's aorta by feeding cholesterol.

Although each school of thought at its inception believed the evidence which it presented was the primary and sole cause of the atherosclerosis, in the later development all in turn have conceded that the secondary factors were of utmost importance. What is primary and what is secondary in the evolution of atherosclerosis has not been definitely proved, but it is encouraging to know that there are several components and that some of these may be influenced.

Accrued from present knowledge, atherosclerosis depends on the following constituents:

1. Cholesterol metabolism
2. Physical and chemical changes of the aortic wall as influenced by the blood pressure, age and intoxication
3. Constitutional or hereditary disposition

Which of the foregoing factors are primary and which are secondary, which can be influenced and which cannot be, are questions that must be explored before any hope of the alleviation of atherosclerosis can be entertained.

With the pendulum swinging toward the belief that atherosclerosis depends on cholesterol metabolism, this possibility in the pathogenesis should be exploited, as it is a factor that may be partly controlled.

The scope of this work embraces only atherosclerosis of the aorta and the immediate factors that influence it. What determine the latter will not receive serious consideration.

The infectious type of lesion (syphilis and rheumatic fever), calcification of the media of the muscular arteries (Mönckeberg's sclerosis), periarteritis nodosa, necrosis of the media (Erdheim), arteritis obliterans, involution sclerosis (ductus Botalli, umbilical cord, vessels of the female organs) and pure senile sclerosis have ostensibly a dissimilar etiology (Jores and Kaufmann) and will not be discussed.

## I. PRELIMINARY DETERMINATIONS

### A. THE FAT CONTENT OF THE AORTA IN 500 CASES OF THE WHITE AND THE COLORED RACE

Because recent experimental, pathologic and clinical observations have indicated a possibility that the cholesterol metabolism may influence the development of atherosclerosis of the aorta, every effort should

be made to verify or disprove this assumption, for the universally accepted notion is that this process is inevitable and the consequence of senescence.

The literature contains a multitude of reports correlating atherosclerosis with diet, blood pressure, infections, environment, climate, race, etc. These communications have in many instances been contradictory because of the lack of uniformity in the methods of examination and also because of the diversity of source.

In the majority of the accounts the diagnosis of atherosclerosis is based on the blood pressure and clinical examinations. That hypertension predisposes to atherosclerosis is granted, but the two conditions are not synonymous and may exist independently. The diagnosis of atherosclerosis of the aorta by examination of the peripheral vessels (rolling them between one's fingers—Stocks) is misleading, as the association of atherosclerosis of the aorta with atherosclerosis of the peripheral vessels in the early stages of the disease is uncommon. Indeed, when the peripheral vessels are affected, the aorta is usually spared (Jores). With present methods of examination the diagnosis of atherosclerosis of the aorta is impossible except when the condition is far advanced (x-ray pictures and Lange's capillary test).

The nonconformity of the statistics have confused what little there was known, and many avenues of research have been discontinued and the workers discouraged because of this. That an application of experimental data to man can be reached only through statistical studies is easily understood. Such information can be accepted only if the investigators conform to a standard method of examination.

As clinical methods for the early recognition of atherosclerosis are still unsatisfactory, it follows that postmortem evidence is necessary. But again difficulties arise in that different pathologists evince varying opinions as to the severity of atherosclerosis. What is considered as severe atherosclerosis in Freiburg, e. g., is adjudged as moderate to slight atherosclerosis in Chicago. Moreover, diffuse atherosclerosis, whether focal or generalized, may be overlooked if the intima is not appreciably altered grossly.

Standard criteria were consequently left to be desired until Aschoff noted that the doubly refractive bodies in atheromatous aortas might be cholesterol esters, and he proved it chemically through the work of Windaus. Unfortunately, the detailed results of the latter cannot be accepted because he failed to remove the adventitia of the aorta and thus included large quantities of fat that was not related to the disease process. It was Schönheimer who realized this error and who by repeating the work of Windaus, removing the adventitia beforehand, found that the total amount of extractable fat in the aorta was directly proportional to the degree of atherosclerosis. Moreover, the lipoid was

mainly composed of free cholesterol and its ester, and in atherosclerotic aortas the relationship between these elements was constant, viz., 25 and 57 per cent, respectively.

The analyses of Schönheimer, because of their extensive chemical nature, cannot be used as a routine procedure. But as the proportions of the fatty constituents remain constant (in atherosclerosis) a determination of the total fat content should suffice. The elaboration of this principle was suggested as a problem for research by Dr. Schönheimer.

The material consisted of 500 aortas of adults over 25 years of age, male and female, of the white and the colored race. The autopsies were performed at the Cook County Hospital in Chicago, and the analyses were carried out in the chemical division of the Pathologic Institute in Freiburg, Germany.

*Method.*—The aorta was cut as close to the valves as possible and at the bifurcation of the aorta. All the branches were cut flush to the surface of the aorta. With a pair of forceps, or better still, with the finger-nails, the adventitia was separated at one point from the media, the line of cleavage being found. Once this separation was brought about, the stripping was simple, as the adventitia, especially in atheromatous aortas, separated very readily from the media. Small muscle fibers of the media remained attached to the adventitia, but the amount of fat contained therein is negligible (Schönheimer <sup>c, d</sup>).

The aorta was then chopped in very fine bits by means of a meat chopper or a specially constructed semicircular blade about 20 cm. in diameter with wooden handles on both ends. A hard wooden board with elevated border was used to prevent the loss of any of the material. The finely chopped mass was then placed in a mortar and ground to a fine dry powder with a minimal amount of anhydrous sodium sulphate.<sup>1</sup> The dried aorta was placed in an extraction bottle and a measured amount of carbon tetrachloride (double-distilled) was added so that the fluid extended at least 1 cm. above the solid matter after a thorough shaking. It was safer to use more extraction fluid than less. The extraction was allowed to continue for from four to twelve hours with intermediary shakings every fifteen to thirty minutes. The fat-containing fluid was filtered off, and in a previously weighed evaporating dish a measured quantity of this fluid was evaporated on a water bath. After complete evaporation, the dish was placed in an electric oven at 120 C. for one hour. The dish was weighed again after it had been allowed to cool in a desiccator. The process of drying and weighing was continued until the weight was constant. The amount of fluid used as solvent being known, as well as the amount evaporated and the weight of fat contained in the latter, the total fat content of the aorta was accordingly determined.

*Results.*—Table 1 represents the average amount of fat in the aorta for each of the age groups indicated. Included in this table are both

1. As the material was gathered in Chicago and the extractions made in Freiburg, Germany, it was necessary after drying with sodium sulphate to remove all moisture in a desiccator. The dried substance was wrapped in ether-extracted cotton cloths and preserved in sealed tin cans on ice. These cans, hermetically sealed, were shipped to Europe. The aortas remained perfectly dry and showed no sign of deterioration.

races and sexes. A progressive increase of fat with age is noted, and when these values were plotted (fig. 1a) the curve thus formed approached almost a straight line. The slight deviations could be accounted for by possibilities of error due especially to incomplete extraction.<sup>2</sup>

The heterogeneousness of the group prevented generalization, and in table 2 the two races were separated. Here again an increased fat content of the aorta was noted with age for both races. Between 25 and

TABLE 1.—*The Average Amounts of Fat in the Aorta for All Cases*

Age	Cases	Fat, Gm.	Age	Cases	Fat, Gm.
25-30	42	0.106	51-60	95	0.503
31-40	86	0.185	61-70	67	0.833
41-50	117	0.355	71-	45	1.162

TABLE 2.—*The Average Amounts of Fat in the Aorta for Each Race*

Age	White		Colored	
	Cases	Fat, Gm.	Cases	Fat, Gm.
25-30	15	0.064	27	0.128
31-40	44	0.155	42	0.218
41-50	81	0.366	36	0.372
51-60	13	0.518	25	0.485
61-70	57	0.831	10	0.845
71-	38	1.122	7	1.382

TABLE 3.—*The Average Amounts of Fat in the Aorta for All Cases in Each Sex of Both Races*

Age	White				Colored			
	Male		Female		Male		Female	
	Cases	Fat, Gm.	Cases	Fat, Gm.	Cases	Fat, Gm.	Cases	Fat, Gm.
25-30	5	0.077	10	0.053	8	0.120	19	0.132
31-40	24	0.187	20	0.117	26	0.214	16	0.222
41-50	52	0.351	29	0.366	21	0.210*	15	0.541
51-60	52	0.511	21	0.535	15	0.424†	10	0.618
61-70	40	0.805	17	0.893	8	0.993‡	4	1.036
71-	25	1.086	13	1.180	5	1.289§		

\* When syphilitic aortitis with atherosclerosis was included—0.266 Gm.

† When syphilitic aortitis with atherosclerosis was included—0.467 Gm.

40 years the amount was greater for the colored race. The plotted values (fig. 1b and c) formed curves similar to each other and to that of the total group.

Further division according to sex (table 3) revealed that the white male presented higher values than the white female up to 40 years, at

2. Schönheimer allowed for an error of 10 per cent under 0.06 Gm. of fat and an error of 4 per cent over 0.1 Gm. However, he extracted his aorta for one hundred and forty-four hours. The percentage of error for each individual case in the present work would be much greater, as the extraction was carried out only for from four to twelve hours. Considering the large number of cases employed, the sum total of error naturally becomes much diminished.

which time the latter surpassed the former. In the colored race the female superseded the male throughout when sufficient cases were available. In comparing the two races, the colored male on the average had more fat in his aorta than the white male, except between the ages of 41 and 60 years. This was accountable for by the fact that syphilitic

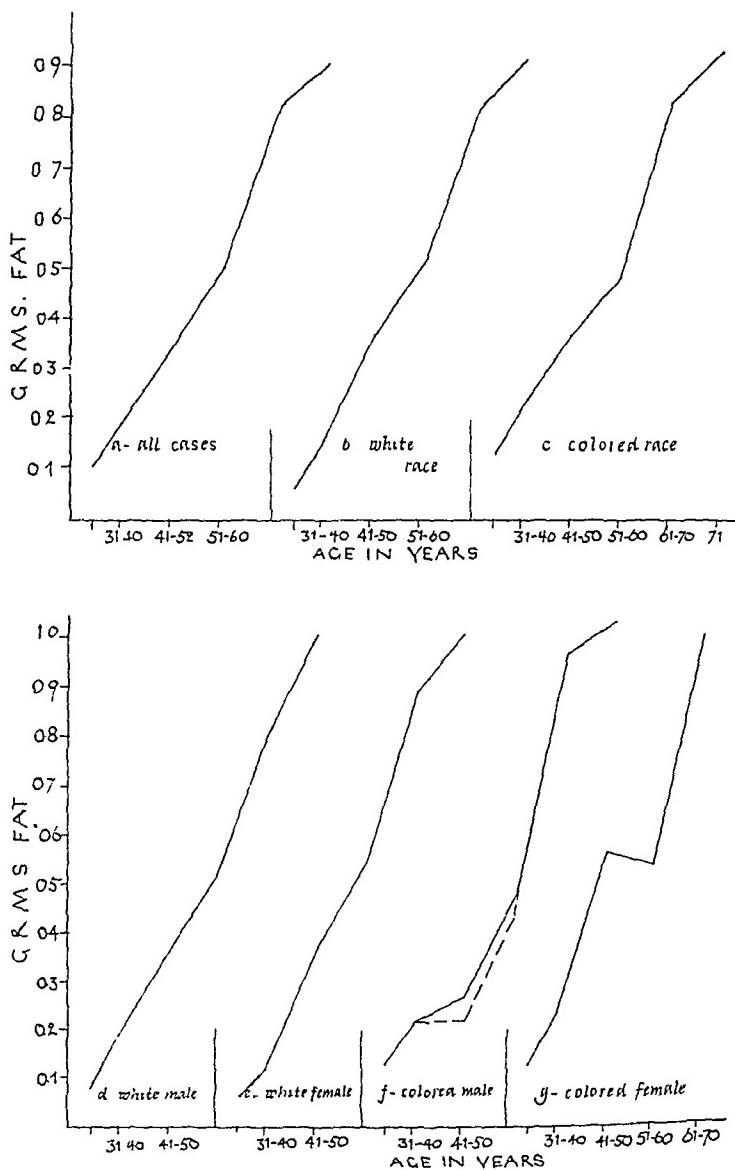


Fig. 1.—Curves of the average amount of fat in the aorta in different age groups of both races and of each race and of the two sexes of each race.

aortitis, whenever present, was excluded in the foregoing analyses. As the colored male at the Cook County Hospital has a higher incidence of syphilitic aortitis (22.02 per cent—Jaffé<sup>a</sup>) as compared with the white male (7.43 per cent—Jaffé), the exclusion of these cases naturally eliminated cases with atherosclerosis. When such aortas were included the average fat values rose.

The colored female in general showed a higher fat content of the aorta than the white female (more detailed and error-corrected comparisons will be given in part C).

Figure 1 *d, e, f* and *g* tenders the curves for the sexes in both races. For the white male and female, whose averages embraced the largest number of cases, the curves approached straight lines. For the colored male a wide bowing was noted between 41 and 60 years and was accounted for by the exclusion of cases with syphilitic aortitis.<sup>3</sup> The insufficient number of cases of colored females accounted for the irregularity of the curve.

*Summary.*—The amount of fat in the aorta increased with age, and when a sufficient number of cases in a homogeneous group were available, the ascent approached a straight line.

#### B. THE RELATIONSHIP BETWEEN THE QUANTITY OF FAT IN THE AORTA AND ATHEROSCLEROSIS

As has been stated in the previous part, the work of Aschoff, Windaus and Schönheimer strongly suggested that the amount of fat in the aorta was parallel to the degree of atherosclerosis. Because their work was carried out on a limited scale, no far-reaching conclusions could be made.

It is necessary to establish definitely such a relationship, as knowledge of the fat content of the aorta would mean little if it did not have a direct bearing on atherosclerosis.

In order to determine this relationship a set of tables was constructed in which the fat contents of the aortas of individuals were subdivided according to sex, age, race and severity of atherosclerosis from a macroscopic standpoint. It may be added at this point that the macroscopic examinations were made close at hand before the aortas were prepared, and all changes were noted. In this preliminary table, three divisions were made, viz., "smooth aortas," "aortas showing slight to moderate atherosclerosis" and "aortas showing moderate to severe atherosclerosis."

Under "smooth" were considered all aortas in which the intima was practically smooth and included small yellow stripes and plaques in almost all cases. Under "slight to moderate atherosclerosis" were included aortas showing discrete node formation of atheromatous or hyaline character and an occasional calcific plaque or ulceration. "Moderate to severe atherosclerosis" embraced all the remaining aortas.

3. Not all cases with syphilitic aortitis were analyzed, but with the addition of a portion of these cases, the curve does not approach that of the white male. This tends to show that syphilitic aortitis and atherosclerosis of the aorta are not related.

In this table many discrepancies were found which could be explained by two possibilities: First, aortas showing local or diffuse atherosclerosis, in which intimal changes are often very slight, may have been included under "smooth aortas." This was particularly common in malignant and, to a less extent, benign hypertension and nephritis. On the other hand, senile ectasia was sometimes misleading and interpreted as severe atherosclerosis. Second, the borderline cases were exceedingly difficult to separate.

To overcome both of these difficulties in part, the mean high and low values were determined for each age group, sex and race, and a new set of tables was constructed in which only these mean fat values were considered, no consideration being made of the gross description. Under this division there were very few discrepancies, although the average fat values for each group remained unchanged. Thus one has a table interpreting atherosclerosis in terms of the fat contents, although the gross description formed the basis of this division.

TABLE 4.—*Fat Values for Smooth Aortas*

Age	Fat Limits	White				Colored			
		Male		Female		Male		Female	
		Cases	Fat, Gm.	Cases	Fat, Gm.	Cases	Fat, Gm.	Cases	Fat, Gm.
25-30	0.02-0.15	5	0.077	10	0.059	7	0.057	14	0.092
31-40	0.03-0.15	15	0.081	17	0.079	15	0.093	9	0.092
41-50	0.06-0.19	26	0.189	10	0.131	14	0.117	4	0.099
51-60	0.06-0.21	14	0.152	7	0.132	4	0.138	2	0.112
61-70	0.06-0.22	3	0.182			2	0.197	1	0.164
71-									

Of course, the interpretation of the intensity of the atherosclerosis was an individual interpretation and might not coincide with that of other authors. But, as the fat content and the severity of the atherosclerosis of the aortas corresponded so closely, this table may serve as a standard of comparison with values from other localities.

In the 179 cases considered under "smooth aortas" (from 0.02 to 0.22 Gm. of fat according to age) there were 6 discrepancies, an error of 3.3 per cent (table 4). These cases showed grossly an occasional nodule and in no instance an outspoken atherosclerosis. When it is considered that 5 of these cases were in persons aged 48 years and over, the possibility that the alterations were senile or that the fat might have been replaced by hyalin is great. In any case, the percentage of error is practically negligible.

Of the 170 cases of atherosclerosis classified as "moderate to severe" (0.16 to 0.8 Gm. of fat according to age—table 5) there were 17 in which the gross description corresponded to "smooth," a 10 per cent discrepancy. These aortas were found in cases of malignant or benign

hypertension and of nephritis. The later was the result of a diffuse atherosclerosis being overlooked on gross examination.

Under "moderate to severe atherosclerosis" (over 0.45 Gm. of fat—table 6) there were no cases in which gross evidence of atherosclerosis was lacking.

The discriminations between the borderlines of slight to moderate and moderate to severe atherosclerosis were more difficult to ascertain, as a definite distinction was not possible. For this reason allowance was made for overlapping: thus the separation of slight to moderate

TABLE 5.—*Fat Values for Slight to Moderate Atherosclerosis of the Aorta*

Age	Fat Limits	White				Colored			
		Male		Female		Male		Female	
		Cases	Fat, Gm.	Cases	Fat, Gm.	Cases	Fat, Gm.	Cases	Fat, Gm.
25-30	0.16-0.44							4	0.215
31-40	0.16-0.44	6	0.213	2	0.169	8	0.269	6	0.338
41-50	0.20-0.5	20	0.340	14	0.325	11	0.305	3	0.205
51-60	0.22-0.6 incl.	26	0.440	40	0.439	10	0.503	5	0.381
61-70	0.22-0.7 incl.	20	0.497	9	0.558	4	0.618	1	0.626
71-	0.23-0.8 incl.	11	0.575	5	0.674	2	0.654		

TABLE 6.—*Fat Values for Moderate to Severe Atherosclerosis of the Aorta*

Age	Fat Limits	White				Colored			
		Male		Female		Male		Female	
		Cases	Fat, Gm.	Cases	Fat, Gm.	Cases	Fat, Gm.	Cases	Fat, Gm.
25-30	0.45 and over					1	0.557	1	0.538
31-40	0.45 and over	3	0.696	1	0.639	3	0.674	1	0.691
41-50	0.5 and over	6	1.308	5	0.951	2	0.716	8	0.855
51-60	0.7 and over	12	1.082	4	1.489	1	0.778	3	1.017
61-70	0.8 and over	17	1.277	8	1.268	2	2.324		
71-	0.9 and over	14	1.486	8	1.511	3	1.718	2	1.614

and moderate to severe. There were 27 cases in the middle group that were grossly interpreted as moderate or severe atherosclerosis, whereas there were but 6 cases in the last group in which a slight to moderate atherosclerosis was present.

Taken all in all, the foregoing classification seemed justified. The percentage of error between the smooth and the atheromatous aortas was almost negligible. It was, after all, this differentiation that was important.

It was of interest to note that in the group of severe atherosclerosis the incidence of ulceration over 41 years was 68 out of 93, or 73 per cent. Yet, in spite of the loss of fat that certainly took place with ulceration, its bearing on the total amount was negligible and this answers possible objections to the classification of atherosclerosis accord-

ing to the fat content of the aorta. Under 40 years the number of cases were too few to allow one to make any generalizations.

Calcification, when severe, may also replace some of the fat, but in a large group of cases this error may be disregarded.

Tables 4 to 6 give the average amounts of fat of the aorta according to the method that has been specified, and it will be noted that in the smooth aortas the amounts of fat were practically similar for both races and sexes. The slight differences could be easily explained by the possibilities of error.

In order to establish a larger number of cases for comparison the fat values of the slight to moderate and moderate to severe atherosclerosis were combined (table 7). In atherosclerosis, the white female up to 50 years of age had a lesser tendency toward fat deposit in the aorta than the white male, and after 50 years she slightly surpassed him. For the colored race the inclination to fat deposit of the female was

TABLE 7.—*Fat Values of All Cases of Atherosclerosis of the Aorta*

Age	Fat Limits	White				Colored			
		Male		Female		Male		Female	
		Cases	Fat, Gm.	Cases	Fat, Gm.	Cases	Fat, Gm.	Cases	Fat, Gm.
25-30	0.16 and over					1	0.557	5	0.280
31-40	0.16 and over	9	0.363	3	0.332	11	0.339	7	0.389
41-50	0.20 and over	26	0.563	19	0.484	10	0.474	11	0.702
51-60	0.22 and over	38	0.643	14	0.736	16	0.550	8	0.619
61-70	0.22 and over	37	0.856	17	0.893	6	1.207	1	0.626
71-	0.28 and over	25	1.086	13	1.189	5	1.289	2	1.614

always greater than that of the male (when sufficient cases were had for comparison). The colored race had an appreciable amount of fat in their aortas from 25 to 30 years, much higher than that of the white race. The colored female presented the highest values up to 50 years of age (cf. part C for complete comparisons).

In a large group of cases, as has been shown, the amount of fat in the aorta ran parallel to the degree of atherosclerosis. These findings verify the suggestions of Schönheimer, whose work was carried out on a small scale. One has then a simple method of comparison that can be used the world over.

What are the advantages of this method over the macroscopic and the microscopic one? From a macroscopic standpoint it is impossible to measure the amount of atherosclerosis as one does an aneurysm, for example. Further, few atherosclerotic plaques in the aorta of an old person might pass as normal and the condition be recorded as a nonatherosclerotic state (considering his age). On the other hand, discrete plaques in a young person might be considered as atherosclerotic. Finally, not all pathologists show the same interest in certain pathologic

states. Considering, then, that most statistics are compiled as after-thoughts, the discrepancies are great.

Microscopic sections are also misleading when the aorta is considered as a whole, as sections are invariably made of the affected areas. Serial sections of the aorta are out of the question when considered as a routine procedure. On the other hand, the simple chemical procedure described gives one a complete picture at once and, in certain respects, is far superior to the serial sections. In the former, one is dealing with a numerical quantity that can be compared with mathematical precision while in the latter, one is dependent on the interpretations of the observer. It is only by adopting such a method that accurate scientific comparisons can be made.

*Comment and Summary.*—The fat content of the aorta, although increasing with age, was found to be directly proportional also to the severity of the atherosclerosis. The facts that the fat deposit was more intense in the young adults of the colored race and that there were variations with sex speak against age as the only determining factor. Because the correlation between the fat content and the atherosclerosis of the aorta in a large group of cases is so precise, it is suggested that the former is not merely a sequence of the latter but that the two are closely dependent on each other. In other parts of the body, e. g., degenerative processes, fibrosis and hyalinization are not accompanied by a corresponding deposit of fat.

#### C. THE INCLINATION TO ATHEROSCLEROSIS OF THE AORTA AS DETERMINED BY THE FAT ANGLE OF THE AORTA (F.A.A.)

In figure 1 *d* and *e*, in which a large homogeneous group came under consideration, the increase of fat in the aorta followed an almost straight course. It was conjectured that if the angle of this inclination could be measured, a simple method of comparing atherosclerosis of the aorta would be had. In order to further this hypothesis, a common scale was adopted so that comparisons could be made with other authors. The scale chosen was similar to that employed in plotting the values in figure 1, as the angles thus formed were of appreciable size and easily measurable. In this graph each millimeter was considered as 0.01 Gm. of fat and also as 1 year. The calculation of an angle may be made by the formula:

$$\text{tangent of an angle} = \frac{\text{altitude}}{\text{base}}$$

in which the altitude is equal to the gram of fat and the base to the age in years. A correction of this formula was necessary because the fat values between 25 and 30 years may vary so that a fixed point was placed at 15 years at which time the fat content is usually constant (about

0.2 Gm.—Schönheimer). Atherosclerosis if present at this age is a rarity. The complete formula is

$$\text{tangent of the angle} = \frac{(\text{grams of fat in the aorta} \times 100) - 2}{\text{age} - 15}$$

With this formula the angles were determined for the various age groups (the groups were considered as a whole, since the larger the group the less was the possibility of error). The mean of all the angles was designated, and this value was considered as the "fat angle of the aorta" (F.A.A.), or the inclination to atherosclerosis of the entire group. The advantage of this method of calculation is that it embraces at once the severity of the atherosclerosis and the incidence.

TABLE 8.—*The Fat Angle of the Aorta (F.A.A.) for All Cases*

Age	Fat, Gm.	F. A. A., Degrees
25-30.....	0.106	24.3
31-40.....	0.185	39.6
41-50.....	0.355	48.2
51-60.....	0.508	51.4
61-70.....	0.833	58.4
71-.....	1.162	62.4
Average F. A. A.....		49.1

TABLE 9.—*The Fat Angle of the Aorta (F.A.A.) for Each Race*

Age	White		Colored	
	Fat, Gm.	F. A. A., Degrees	Fat, Gm.	F. A. A., Degrees
25-30	0.064	19.6	0.128	40.8
31-40	0.155	34.0	0.217	44.8
41-50	0.357	48.4	0.372	49.5
51-60	0.518	51.2	0.485	49.4
61-70	0.831	58.4	0.845	58.8
71-	1.122	61.4	1.382	66.5
Average F. A. A.	45.5		Average F. A. A.	51.6

The fat angle of the aorta (F.A.A.) or the inclination to atherosclerosis of the entire group of cases studied was disclosed to be 49.1° (table 8).

The foregoing value applies to the poorest class of people living in Chicago under similar economic conditions for a greater or a lesser number of years. Of the white persons a high percentage of the older adults were foreign-born (Germans, Irish, Italians, Poles, Croats) but had lived in Chicago from prewar times. A later influx has been guarded against by the strict immigration laws. Among the colored persons practically all were southern-born and the duration of their residence in Chicago was as a rule proportional to their age (from 25 to 31 years—six years: from 71 to 80—forty years).

Comparing the two races (table 9), the colored persons in the second to fourth decades of life had a greater inclination to atherosclerosis

than the white persons. From 41 years onward the two races showed no appreciable differences until after 71 years, when the colored race again had a predominant degree of atherosclerosis. These differences were far beyond the margin of error, as between 25 and 30 years the difference was 100 per cent and between 31 and 40 years and over 71 years it was about 25 per cent.

The average fat angle of the aorta (F.A.A.) for the white race was somewhat lower ( $45.5^\circ$ ) than that for the colored race ( $51.6^\circ$ ). Although this difference falls within an error of 15 per cent and may be discounted, the individual group differences were such that one was justified in saying that the colored race had a stronger inclination to atherosclerosis than the white race. The contention of Stocks is similar to this, but he based his conclusions mainly on clinical diagnosis (death

TABLE 10.—*The Fat Angle of the Aorta (F.A.A.) for Each Sex in Each Race*

Age	White				Colored			
	Male		Female		Male		Female	
	Fat, Gm.	F. A. A. Degrees	Fat, Gm.	F. A. A., Degrees	Fat, Gm.	F. A. A., Degrees	Fat, Gm.	F. A. A., Degrees
25-30	0.077	22.9	0.053	14.8	0.120	38.6	0.132	44.3
31-40	0.187	39.8	0.117	25.9	0.214	44.2	0.222	47.4
41-50	0.351	48.5	0.366	49	0.266	39.5	0.542	60.4
51-60	0.501	50.8	0.535	52.8	0.408	48.2	0.518	50.8
61-70	0.805	57.6	0.693	60.2	0.955	61.8	1.036	61.4
71-	1.066	60.5	1.189	62.9	1.289	64.8		
Average F. A. A.....	46.7		44.4			49.5		52.8

certificate statistics). Camac, who is frequently quoted in relation to atherosclerosis of the colored race, reported only on syphilitic aortitis.

A comparison of the sexes (table 10) revealed that the white female had a lesser tendency to atherosclerosis than the white male at the age of from 25 to 40 years, but that after 40 years the difference was slight. For the entire group the male had a slightly greater inclination to atherosclerosis than the female. This variance was within the possibility of error, yet the individual age group differences fell beyond it.

Most authors, in comparing the incidence of atherosclerosis in males and females, do not take into consideration the severity, as the methods available make such determinations impossible. The reports are unanimous as to the predominance of atherosclerosis in the male (Jores, Cramer, E. Kaufmann). With my method both incidence and severity are considered, and although the male showed the greater tendency to atherosclerosis, it was only in the second to fourth decades.

For the colored race the female presented a more obtuse fat angle of the aorta (F.A.A.) up to 50 years, after which time the difference was negligible. When the possibility of error was considered, the only

dissimilarity was between 41 and 50 years, at which time it reached a 33 per cent variation in favor of the female (syphilis of the aorta included). The fat angles of the aorta (F.A.A.) for the male between 41 and 60 years fell below the expected values owing to the exclusion of syphilis of the aorta (cf. earlier paragraph).

TABLE 11.—*The Fat Angle of the Aorta (F.A.A.) for Smooth Aortas*

Age	White			Colored		
	Cases	Fat, Gm.	F. A. A., Degrees	Cases	Fat, Gm.	F. A. A., Degrees
25-30	15	0.065	22.2	21	0.072	24.4
31-40	32	0.080	16.8	24	0.093	21.4
41-50	36	0.137	21.2	18	0.113	17.2
51-60	21	0.145	17.4	6	0.128	15.2
61-70	3	0.182	17.9	3	0.186	18.4
71-						
Average F. A. A.....			19.3	Average F. A. A.....		

TABLE 12.—*The Fat Angle of the Aorta (F.A.A.) for Slight to Moderate Atherosclerosis*

Age	White			Colored		
	Cases	Fat, Gm.	F. A. A., Degrees	Cases	Fat, Gm.	F. A. A., Degrees
25-30				4	0.215	55.2
31-40	8	0.202	42.3	14	0.290	54.3
41-50	34	0.334	46.3	8	0.301	43
51-60	36	0.440	54.5	15	0.468	48.9
61-70	29	0.517	44.9	5	0.645	51.2
71-	16	0.606	44.5	2	0.654	46.5
Average F. A. A.....			46.5	Average F. A. A.....		

TABLE 13.—*The Fat Angle of the Aorta (F.A.A.) for Moderate to Severe Atherosclerosis*

Age	White			Colored		
	Cases	Fat, Gm.	F. A. A., Degrees	Cases	Fat, Gm.	F. A. A., Degrees
25-30				2	0.548	76.8
31-40	4	0.662	72.7	4	0.678	73.1
41-50	11	1.146	75.1	10	0.827	69.8
51-60	14	1.182	71	4	0.957	66.9
61-70	25	1.275	68.4	7	1.859	73.2
71-	22	1.496	67.9			
Average F. A. A.....			71	Average F. A. A.....		

In order to establish a correlation between the fat angle of the aorta (F.A.A.) and the intensity of the atherosclerosis, the degree of inclination for the values in part I B was determined. Because a larger number of cases were desirable for comparison, the sexes of each race were considered as one (tables 11 to 13).

The average fat angles for both races were similar. The smooth aortas had a fat angle of 19.3°, the aortas showing slight to moderate

atherosclerosis an angle of  $48.1^\circ$ , and the aortas showing moderate to severe atherosclerosis an angle of  $71.4^\circ$  (fig. 2).

From these values it was deducted that, for the average, the colored race living in Chicago begins to present a slight to moderate atherosclerosis at from 25 to 30 years, while for the white race the first indication appears between 31 and 40 years. When the races were considered as a whole, the average person over 25 years coming to autopsy at the Cook County Hospital had a slight to moderate atherosclerosis.

*Comment.*—From the evidence offered in part C, the results of parts A and B take shape and form, so to speak. In other words, the fat angle of any group being known, an insight into the incidence and severity of atherosclerosis in the group is obtained.

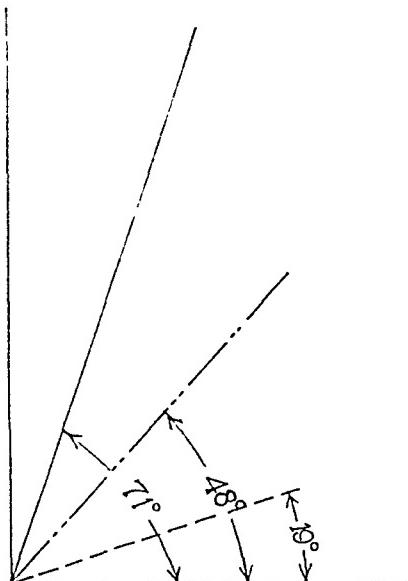


Fig. 2.—The fat angle of the aorta (F.A.A.): Angle of  $71^\circ$ , moderate to severe atherosclerosis; angle of  $48^\circ$ , slight to moderate atherosclerosis; angle of  $19^\circ$ , smooth aortas.

The nature of the curves and the calculation of their angles of inclination seemed to indicate that the accession of atherosclerosis is related only to age. On closer inspection, however, the differences noted with the two races and sexes indicated that other factors were at play. As has already been suggested, the cholesterol metabolism, the physical and chemical changes of the aortic wall and heredity all play their respective rôle. With the standard of measurement presented the influence of these factors can be judged more accurately. It is hoped that with this standard of measurement similar studies will be carried out by other authors, as it is only by exact methods of comparison that definite conclusions can be drawn.

*Summary.*—A simple chemical method has been described to determine the fat content of the aorta. The latter was found to be directly proportional to the severity of atherosclerosis in an examination of 500 aortas.

Because in a homogeneous group the increase of fat in the aorta followed an almost regular progression with age, the inclination of this ascent could be determined by the formula:

$$\text{tangent of the fat angle of the aorta (F.A.A.)} = \frac{(\text{grams of fat} \times 100) - 2}{\text{age} - 15}$$

The value thus obtained was designated as the fat angle of the aorta (F.A.A.) or the inclination to atherosclerosis. The advantage of this method is that it interprets both the incidence and the severity of atherosclerosis when a large group is considered as a whole.

It was found that an angle of 19.3° indicated a smooth aorta, an angle of 48.1° a slight to moderate atherosclerosis, and an angle of 71.4°, a moderate to severe atherosclerosis.

The fat angle of the aorta (F.A.A.) for all cases examined was 49.1° or, translated, a slight to moderate atherosclerosis.

The colored race had a greater inclination to atherosclerosis than the white race, which was most marked between 25 and 40 years. The white race developed atherosclerosis later than the colored race.

The white female had a lesser tendency to atherosclerosis than the white male but only between 25 and 40 years.

The colored race showed no great difference in their F.A.A. with sex except between 41 and 50 years, when the female showed a predominant amount of atherosclerosis.

It was suggested that with the aforesaid standard of measurement the relationship of atherosclerosis of the aorta to the cholesterol metabolism, the physical and chemical changes of the aorta and the heredity may be better understood.

## II. BEARING OF CHOLESTEROL METABOLISM ON INCLINATION TO ATHEROSCLEROSIS OF THE AORTA (F.A.A.)

Knowledge of the cholesterol metabolism is limited. What determines the blood cholesterol is equally obscure. It is customary to speak of oxogenous and endogenous cholesterol.

Although it is generally accepted that the cholesterol of the blood remains constant irrespective of the diet (Bürger, Beumer, Thannhauser) the endogenous cholesterol is indirectly dependent on it. Thannhauser expressed the belief that the endogenous cholesterol in adult human beings is obtained more from the fat depots than by synthesis.

In infants Beumer demonstrated that, notwithstanding the presence of a negative cholesterol balance, the cholesterol of the brain increases twelvefold in the first eighteen months. In herbivorous animals the synthesis of cholesterol in the body has been more definitely established.

Schönheimer has shown in rabbits that the vegetable sterols (phytosterol) are not absorbable, yet cholesterol is present in the rabbit's blood.

Whatever the relationship between the endogenous and exogenous cholesterol may be, accurate studies seem to indicate that the former is to a great extent dependent on the latter, although the influence may not be a direct one (Bürger, Beumer, Thannhauser,<sup>b</sup> Schönheimer<sup>a</sup>). The term "endogenous cholesterol" as used in this paper refers to that cholesterol not derived directly from the nutrition.

#### A. EXOGENOUS CHOLESTEROL

If any relationship could be established between the diet, especially as it affects the cholesterol metabolism, and atherosclerosis, a great stride forward would have been made toward the solution of the pathogenesis and prevention of the disease.

From an experimental standpoint the production of atheromatous lesions in the aorta of the rabbit by feeding cholesterol dissolved in oil was first shown by Anitschkow and Chalatow and verified by numerous others (Wacker and Hueck, Bailey, Versé, Schönheimer,<sup>a, b, c</sup> Thölldte, Rosenthal).

The objections to comparing the experimental lesions to those found in man are that a severe hypercholesterolemia is produced in the rabbit, rarely found in man, and that not only the aorta but the pulmonary artery, as well as the veins (Schönheimer<sup>b</sup>) and other organs, show deposition of cholesterol. The lesions in the experimental animals supposedly represent only the lipoid deposits seen in nursing infants, at puberty, etc. (Zinzerling,<sup>a</sup> Aschoff,<sup>b</sup> Klotz and Manning).

Many objections of the types stated have been overruled by further experiments. Thus Anitschkow,<sup>c</sup> by feeding rabbits diluted milk and egg yolk over a period of two and a half years, found a mild type of atherosclerosis with only a slight elevation of the blood cholesterol and slight to no deposits in other organs. Similar results were obtained by Chuma and Zinzerling<sup>b</sup> who fed hydrous wool fat to rabbits over a long period.

The relationship of the fatty streaks and plaques in youth to atherosclerosis has also been much disputed. Virchow believed them to be separate processes (also Ribbert, Klotz and Manning, Beitzke, Lubarsch, E. Kaufmann), but the recent thorough work of Zinzerling,<sup>a</sup> who stained human aortas of various ages *in situ* with Sudan III, has shown that these fatty deposits are found in the same locality as the atherosclerotic ones, and that the two processes are definitely connected (Aschoff,<sup>c, e, f</sup> Anitschkow<sup>c</sup>).

The foregoing comparison was not given with a view to showing that experimental and human atherosclerosis are similar, as there are many differences indeed. Anitschkow<sup>c</sup> himself showed that in experimental

atherosclerosis the elastic lamellae are never infiltrated as in man, and that the hyaline plaque and scar formation, as well as the ulceration, are rarely seen in the experimental animal. What is stressed here is that the similarities which are present are of such a character that one must admit that cholesterol (especially the esters) plays some rôle in the pathogenesis of atherosclerosis.

In dogs and other carnivora the enteral administration of cholesterol does not produce atherosclerosis, especially in young animals (Anitschkow<sup>b</sup> Kawamura, Cirio, Yuasa, Adler, Tsunoda and Umehara). This is because carnivorous animals excrete their cholesterol very rapidly (Rothschild) in contradistinction from herbivorous animals, which retain it. Only when the cholesterol can be fixed in the blood do atheromatous lesions occur (as after castration—Murata and Kataoka,<sup>a</sup> Löwenthal). This does not disprove that enteral cholesterol may produce atherosclerosis, as the experiments on dogs have been carried on for relatively only a short time (two years—Tsunoda and Umehara), and a type of atherosclerosis does occur spontaneously in old dogs (Zinzerling,<sup>b</sup> Krause).

In man the same disposition to excrete cholesterol exists as in other carnivorous animals. Many authors believe that enteral cholesterol does not affect the cholesterol content of the blood at all. Ssokoloff obtained no digestive hypercholesterolemia in normal men after administering 3 Gm. of cholesterol dissolved in oil daily for three days. His examinations of the blood were made after twenty-four hours. Mjassnikow obtained negative results in normal men after feeding 2 Gm. of cholesterol in the form of eggs or their equivalent. He examined the blood of his patients from two to three hours after feeding, using a colorimetric method of determining the cholesterol. In a similar way, Rouzaud and Cabanis obtained no digestive lipemia. However, the methods employed by the aforementioned authors were faulty, either in the amount of cholesterol fed, the time after feeding at which determinations of the blood cholesterol were made, or the methods used for determining the cholesterol.

Bürger uses the following method of examining his patient:

1. At least 5 Gm. of cholesterol dissolved in 100 cc. of oil at body temperature must be given on a fasting stomach.
2. The determinations of the blood cholesterol should be carried out at four, eight and twenty-four hour intervals.
3. The Windaus digitonin method for determining blood cholesterol must be employed, as colorimetric methods may obscure the results, especially if the serum is highly colored.

Following this method, Bürger found that in all adults examined the blood cholesterol increased over 100 per cent in four hours; the relationship of free cholesterol to cholesterol esters remained constant. In

eight hours the blood cholesterol was 50 per cent above normal, and in twenty-four hours it was normal. Positive results were obtained by Widal, Weil and Laudot and Arndt.

Barreda from Thannhauser's clinic recently repeated Bürger's work, but no determinations were made after eight hours. In normal persons he found an increase of free cholesterol of from 7 to 25 per cent (in 5 of 6 cases) and an increase of total cholesterol of from 6 to 17 per cent (in 4 of 6 cases). These readings were after four hours.

It follows then that there is a distinct digestive lipemia in man, and that its peak is in four hours. Considering that as a rule one eats every four to five hours, it may be inferred that, depending on the type of food eaten, an almost constant lipemia may be present during the day.

A large part of the exogenous cholesterol passes through the body of the carnivorous animal, although it is rapidly excreted. The purpose of the cycle of intestine to thoracic duct, to blood, to liver, to bile, to intestine is not understood. As will be shown later, atherosclerosis of the aorta as it occurs in man is never found in animals which are fed a diet devoid of cholesterol and which must synthesize their own sterols. Can one assume that exogenous cholesterol, although not to be differentiated from the endogenous type, may nevertheless have singular properties not yet recognizable?

If, on the basis of the foregoing hypothesis, the atherosclerosis may be influenced by the type of cholesterol, and the latter in turn is partly determined by the diet, an avenue is opened for a research that should be encouraged.

The first large stride in that direction has been taken by Raab<sup>b</sup> who, by personal information and through the literature, has gathered all available data concerning the interconnections of diet and atherosclerosis. Table 14 is a compilation of his results as well as of additional information obtained in the present investigation.

On scanning this table one notices that the accounts are based, to a great extent, on clinical observations, and that emphasis is placed on protein diet and elevation of blood pressure. This may be explained by the evidence of the earlier workers on experimental atherosclerosis who utilized animal tissues (Ignatowsky, Steinbiss and Stuckey). It was then thought that the high protein content was the active principle, but it was later proved that the cholesterol component was the important one (Stuckey, Anitschkow,<sup>a</sup> Chalatow,<sup>a</sup> Wacker and Hueck). Similarly, the blood pressure was thought to be influenced by a high protein diet. There have been many studies made disproving this conception, the most striking being that of Thomas. He found that Eskimos, who live mainly on red meat, have an average blood pressure between 40 and 60 years of 129 systolic and 76 diastolic. This pressure is much lower than that found in the United States (Gager, Foster).

TABLE 14.—*The Relationship of Diet and Blood Pressure to Atherosclerosis*

Race	Diet			Blood Pressure, Mm. of Mercury	Incidence of Atherosclerosis	Comment	Authors and References
	Protein	Fat	Carbo-hy-drate				
1 White, Europe	Low, no meat or eggs	Low, no butter, little milk	Moderate	75% under 120	Not given	Monks who rarely talk	Salle
2 White, Europe	High, meat and eggs	High, butter and milk	High	70% over 120 between 50 and 90 yrs., 140-160	Not given	Monks who go among their people	Salle
3 White, Germany	Low, very little fat of any kind	Low	Moderate	.....	Much decreased	During and shortly after the war	Aschoff (f)
4 White, Vienna, Austria	Normal European diet			.....	40% of autopsies in older persons, up to 88%	1921-1923	Schubert, F.: Klin. Wchnschr. 31 : 751, 1925
5 White, Eng-land	Normal European diet			140/90 at 60 yrs.	High	Clinical and post mortem	Donnison Roger
6 White, S. Amer-ica	Very high, meat 30% of diet, also eggs 85 Gm. per day	High	Very high (?)	High (?)	Clinical and post mortem	Castex, S.: La hypertension arteriale, Buenos Aires, H. Andretta, 1929	
7 White, New York	High	High	High	23% over 150 at 40 to 60 yrs.	High	Private (clinical) practice	Gayer
8 White, U. S. A.	High	High	High	High in 5 million out of 120 million	High	Clinical	Mayors, quoted by Castex
9 White, U. S. A.	High	High	High	High	40 yrs., 11.4% 50 yrs., 26.3% 60 yrs., 30%	From death certificates (mostly clinical)	Stocks
10 White, U. S. A.	Average American diet			Average American	20-29 yrs., 3.14% 30-39 yrs., 9.14% 40-49 yrs., 26% 50-59 yrs., 48% 60-69 yrs., 78% 70 yrs. and over, 90%	Postmortem records	Ophüls
11 White, U. S. A.	Average American diet			45% over 130 22% over 140	Negligible	Freshman class Univ. of Calif.	Alvarez, Wlezen and Mahoney
12 White, U. S. A.	Normal American diet			16% over 140 (1923) 90% over 140 (1924)	Negligible	Freshman class, Univ. of Michigan	Diehl and Sutherland
13 White, U. S. A.	Normal American diet			10% over 140	Negligible	Freshman, Harvard University	Palmer
14 White, Eskimos	High, red meat and liver	Low	Low	40 to 60 yrs., 129/76	Negligible	Clinical diagnosis, diet optimum	Thomas
15 White, Egypt	High	High	High	35 to 55 yrs., 10% hypertension	Frequent	Private practice, clinical	Ismail
16 White, Egypt	Low	Low	High	No hypertension	No atherosclerosis	Hospital practice, poor class	Ismail

TABLE 14.—*The Relationship of Diet and Blood Pressure to Atherosclerosis—Continued*

Race	Diet		Carbo-hydrate	Blood Pressure, Mm. of Mercury	Incidence of Atherosclerosis	Comment	Authors and References
	Protein	Fat					
17 Brown, Indians, Calcutta	Low	Low	High	Lower than Europeans	Similar to Europeans	Postmortem material	Roger
18 Brown, 7.6% Indians, British	7.6%	1.4%	91%	Low	50% lower than in Europe	Eggs, milk, butter in background; i.e., little of eggs, milk and butter; many cereals (poor class)	McCarrison and Raab (b) quoted by Raab (b)
19 Brown, 9.4% Indians, British	9.4%	10.6%	80%	High	.....	Well to do class	Raab (b)
20 Brown, Malay, Dutch East Indies	Low	Low	High	Low	Very seldom	Blood cholesterol as well as calcium low	de Langen
21 Yellow, Kirgisen-Steppe, 10-20 lbs. of goat meat	Very high, 10-20 lbs. of goat meat	High (mare milk)	High	.....	High	Apoplexy and contracted kidneys common	Kucsynski
22 Yellow, 9% Chinese	9% (plant)	4% (plant)	87%	5 to 10 lower than white	Infrequent	Clinical	Tung (b) Maxwell, quoted by Raab (b)
23 Yellow, Low Chinese	Low	High	Among patients 20 over 160	Among 4,000 patients 20 over 160	Infrequent	Clinical	Foster
24 Yellow, Low Chinese	Low	High	Low	Among 1,924 patients 50 with atherosclerosis	Clinical	Personal communication of Dienarde with Raab	
25 Yellow, 13.7% Japanese	4.9%	81.4%	Relatively low	Diseases of arteries, 21.8%	Statistics from insurance company	Rubner, quoted by Raab (b)	
26 Black, East Africa	Low	Low	High	60 yrs., 105/67	Negligible	Clinical and post mortem	Donnison
27 Black, Jamaica	Moderate	Moderate	High	33% over 140	Frequent	.....	E. T. Newmann
28 Black, U. S. A.	.....	.....	.....	.....	40 yrs., 14.8% 50 yrs., 31.0% 60 yrs., 45.0%	Death certificate diagnosis	Stocks

Increased blood pressure definitely predisposes to atherosclerosis, but one cannot imply that the two conditions are concomitant. (See Alvarez, Wiezen and Mahoney, Diehl and Sutherland, and Palmer in table 14 for reports on freshman university students.) Thus, in spite of the large material gathered, accurate deductions cannot be made. Yet there are many observations which suggest that precise information would be of great value, and that further work along similar lines but with a definite standard of measurement should be encouraged.

One of the outstanding features of table 14 is that in no race for which a high cholesterol intake (in the form of eggs, butter and milk)

and fat intake are recorded is atherosclerosis absent (America, United Kingdom, Central Europe, South America, Mongolia: Majors, Rogers, Schubert, Castex, Kuczynski, respectively). Where a high protein diet is consumed, which naturally contains small quantities of cholesterol, but where the neutral fat intake is low, atherosclerosis is not prevalent. Thus, in the Japanese race, by which a diet rich in protein but low in neutral fat (4.9 per cent compared with 11.4 per cent in the western diets—Rubner) is consumed, the incidence of atherosclerosis is much lower than it is in the United States, e. g. (3.1 to 21 per cent, respectively—Rubner). Similarly, the Eskimos who, contrary to current opinion, eat very little neutral fat (as the oils extracted from the liver, etc., are used for heating, lighting and cooking—Thomas) have a low incidence of atherosclerosis, although their diet consists mainly of red meat (Thomas).

As mentioned by Hoppe-Seyler in 1857 and verified by Versé,<sup>a</sup> Schönheimer,<sup>a</sup> Thannhauser, Bürger and others, the neutral fat paves the way for cholesterol absorption. Diets high in cholesterol and low in neutral fat may result in a much lower blood cholesterol than a diet high in neutral fat and low in cholesterol. This has been proved definitely experimentally (Versé, Wacker and Hueck). Aschoff<sup>t</sup> traces the marked decrease of atherosclerosis in Central Europe following the war to the low intake of fat.

Of interest is the report of Ismail in Egypt, who has communicated that among his private patients, whose diet is similar to that of the Europeans, the incidence of atherosclerosis is high, while in his hospital practice composed mainly of natives, who subsist largely on a carbohydrate diet, the incidence of atherosclerosis is low. Here one has people living under similar conditions of climate and environment, but consuming different foods, with a marked variation in their predisposition to atherosclerosis.

Saile compared monks living on an absolute vegetarian diet (without meat, eggs or butter) with another group of monks who were non-vegetarian, subsisting on a diet similar to that of the average European. Unfortunately, his results dealt mainly with blood pressure; he showed that only 25.4 per cent of the vegetarians had a systolic blood pressure over 120 mm. of mercury. In comparison, 70.4 per cent of the nonvegetarians had a blood pressure over that amount. He inferred a similar relationship to arterial changes. It must be stressed at this point that the vegetarians usually considered in the literature abstain from meat but consume eggs, milk and butter to a high degree. This diet, high in cholesterol and neutral fat, may account for the contradictory opinions.

One discrepancy was reported by Roger, who found that the incidence of atherosclerosis in Bengal Indians was similar to that in England. The Indians eat a diet comparatively low in protein and fat, very similar to that of the southern Chinese, in whom the incidence of atherosclerosis is low (Foster). It is especially in such cases that an accurate method of measurement of atherosclerosis is needed.

In animals the only lesions that resemble human atherosclerosis of the aorta were found in aves, especially parrots (Fox, Wolkoff,<sup>a</sup> Nieberle and Beneke). These birds are meat and seed eaters, and thus cholesterol and neutral fats, respectively, are plentiful in their diet. That other animals also consume meat and fat (gormandizing animals) and rarely develop atherosclerosis is true, as it cannot be denied that age and construction of the aorta (Fox) play a definite rôle, yet it is significant that those who do acquire an atherosclerosis similar to that in man consume cholesterol and neutral fats.

As has been stated, the foregoing statistics cannot be considered as final, as no definite or accurate standard of measurement was employed, but they offer some hope that cholesterol and neutral fat in the diet may influence the inclination to atherosclerosis, and that further work should be carried on in which a common exact method of comparison is used. The method suggested in part I for the determination of the inclination to atherosclerosis is offered as a means to this end.

#### B. A COMPARISON OF THE DIETS OF THE COLORED AND THE WHITE RACE AND ITS BEARING ON THE FAT ANGLE OF THE AORTA (F.A.A.)

The fat angles of the aorta (F.A.A.) for both races and sexes were equal, except for the discrepancy between 25 and 40 years. In seeking for the cause of this difference, several possibilities presented themselves.

The persons of the colored race of Chicago considered in this paper were for the greater part southern-born, and in determining the number of years that each group had resided in Chicago it was found that this was roughly proportional to their age (table 15).

TABLE 15.—*The Average Number of Years of Residence in Chicago of the Colored Race*

Age	Male	Female
25-30.....	6 yrs.	12 yrs.
31-40.....	13 yrs.	12 yrs.
41-50.....	16 yrs.	10 yrs.
51-60.....	22 yrs.	9 yrs.
61-70.....	22 yrs.	27 yrs.
71-.....	25 yrs.	40 yrs.

Thus, up to 40 years for the male and up to 60 years for the female the average residence in Chicago was relatively short. Considering that the colored people of the South are very poor and live in a rather primitive manner, deficiency diseases are exceedingly common there.

In an effort to ascertain the nature of the diet of the southern colored people, a questionnaire was sent out to various Negro institutions of the South. Answers were received from Louisiana, Alabama, North Carolina and Texas.

The general opinion of these reports from the Negroes was that the greatest proportion of their diets (as with all poor people) consisted of carbohydrates (cornbread, potatoes, molasses). To a much less extent meat was consumed, and butter, milk and eggs were eaten in minimal quantities.

When the colored persons came to the North their standard of living was immediately elevated. Although a large proportion of them remained very poor, their labors brought them higher returns, and what they themselves could not supply, society supplied for them. Many of the colored persons entering the Cook County Hospital are comparatively well-to-do, however, and their consumption of meat, milk, eggs and butter approaches and to some extent surpasses that of the white persons interned at the hospital. Although the diet of the foreign white persons had also changed, the difference was much less marked, as these came from Central Europe. Further, the younger white persons (from 25 to 35 years of age) are for the most part American-born.

Can this change of diet and also environment of the colored race play some rôle in their cholesterol metabolism? Whether the increase of cholesterol, neutral fats and proteins in their diets increased directly the cholesterol content of the blood, or whether this change of diet caused a disturbance of the cholesterol metabolism in general cannot be stated (Weiss and Minot). The fact remains that there is an increase of fat deposit in their aortas, and although there are no studies determining the cholesterol content of their blood, the following observation suggests that there might be an elevation.

Joël has shown by simultaneous determinations of the blood cholesterol and examinations of the cornea in young persons (about 25 years of age) that a close relationship exists. Thus he found elevations of cholesterol up to 320 mg. per hundred cubic centimeters of blood in young persons with arcus lipoides. Experimentally arcus lipoides is easily produced in rabbits by feeding them cholesterol (Versé, Kolen, Schönheimer,<sup>a</sup> Rosenthal). This condition in rabbits resembles almost exactly that in young persons.

A comparison of the occurrence of arcus lipoides in persons between 25 and 40 years of the two races revealed that it was not recorded as occurring in any of the white persons, whereas among the colored per-

sons it occurred in 46 per cent of the males and in 36.3 per cent of the females. In only one instance did arcus lipoides occur in a 30 year old colored man who presented an aorta with a few fatty plaques and streaks and a fat content of 0.041 Gm.

Can it be deducted on the basis of Joël's work that the cholesterol content of the blood of the colored race between 25 and 40 years is higher than that of the white race (or at least between 25 and 30 years, for on the basis of the experimental work of Kolen arcus lipoides may remain for years after the disappearance of the hypercholesterolemia)? If so, perhaps the blood cholesterol does play some active rôle in atherosclerosis. Further, can one suggest that a hypercholesterolemia occurs before atherosclerosis as in the case of the 30 year old colored man who had an arcus lipoides and only a few yellow streaks as well as a low fat content in his aorta?

If the blood cholesterol of the colored race is higher than that of the white race between 25 and 30 years, what can account for this difference, since the diets of the two races while living in Chicago are about similar?

Is it possible that the Negro living under rather primitive conditions in the South, where proteins and cholesterol-containing foods are luxuries, is slow, when transferred to a different environment with a higher standard of living, to accommodate himself to the new diet, and a hypercholesterolemia occurs? After a longer residence acclimatization is established, and the cholesterol in the blood returns to the level of the white man.

One is tempted to compare what has been described with what occurs in the cholesterol-fed rabbit, which at first reacts with hypercholesterolemia, but in which, after protracted cholesterol feeding, the cholesterol content of the blood falls to normal (Thölldte, Schönheimer,<sup>a</sup> Rohrschneider).

There is a strong possibility that a disturbance of the cholesterol metabolism of the Negro may have taken place before his emigration to the North. Unfortunately the only evidence that I could find regarding the incidence of atherosclerosis of the colored race in America was based on clinical diagnosis (Stocks), and the statistics showed a preponderance of atherosclerosis in the colored race. Most probably syphilitic aortitis was included in this study, and thus these values cannot be accepted. The anatomic statistics of the earlier authors (Camac) dealt mainly with syphilis.

Bearing in mind that the American colored person took his origin from North Africa not very long ago, one may ask whether this more drastic change of environment may not still be leaving its stigmas on the present colored population of America.

Reports from Africa (East African Negroes) by Donnison show that the East African, living primitively and excluding meat and cholesterol-containing food from his diet almost completely, has an almost negligible incidence of atherosclerosis.

The foregoing evidence is indicative only of the possibility that cholesterol metabolism plays a rôle in atherosclerosis of the aorta and needs further verification. The fact that increased blood pressure, to which the young Negro is susceptible, may also influence the lipoid deposit in the aorta will be discussed later.

#### C. THE RELATIONSHIP BETWEEN ENDOGENOUS CHOLESTEROL AND THE FAT ANGLE OF THE AORTA (F.A.A.)

Experimental and clinical observations strongly suggest that the body synthesizes a portion of its cholesterol (Beumer and Schönheimer<sup>e</sup>). In human adults this fraction is very small or may even be transported from the fat depots (Thannhauser). In spite of the varying amounts of cholesterol obtained from the diet (0.018 to 1.4 Gm., depending on the cholesterol content of the diet—Thannhauser) and the endogenous quota (0.03 Gm.—Thannhauser) the blood cholesterol remains constant. The regulating mechanism is believed to be in the liver (Bürger, Thannhauser<sup>b</sup>).

Hypercholesterolemia from an endogenous origin may be divided into the following groups (Bürger):

1. Hypercholesterolemia of pregnancy
2. Transport hypercholesterolemia
3. Retention hypercholesterolemia
4. Cytolytic hypercholesterolemia
5. Narcosis hypercholesterolemia

As hypercholesterolemia, in order to effect a lipoid deposit in the aorta, must act over a longer period of time, the interest here lies in the second, third and fourth divisions.

*Transport Hypercholesterolemia.*—The hypercholesterolemia of this group is supposedly due to the lipemia from inanition or hunger (Schultze, Thannhauser,<sup>b</sup> Bürger). The theory is that with inanition the fat is transported from the fat depots by way of the blood stream to the parenchyma of the body. In the course of this, the cholesterol present in the fat depots is also carried into the blood. It does not follow that with a given amount of neutral fat there will be a corresponding amount of cholesterol transported. In tuberculosis and carcinoma, e. g., the cholesterol of the mesenteric and subcutaneous fat becomes more concentrated (Wacker) with inanition. If this inanition or hunger is protracted over a longer period, an exhaustion of the lipoid

and cholesterol depots takes place and a hypocholesterolemia may set in, as found in hunger edema (Knack and Neumann; Feigl and Mathies, cited by Bürger).

Under transport hypercholesterolemia Thannhauser included atherosclerosis, diabetes mellitus, chronic glomerular nephritis, tuberculosis and carcinoma. Although there are other factors at work influencing this hypercholesterolemia, such as acidosis in diabetes mellitus, their exact nature is not known.

(a) Atherosclerosis of the Aorta: Hypercholesterolemia in atherosclerosis has been reported (Mjassnikow, Pribram) as well as normal or lower than normal blood cholesterol values (Hunt). The negative reports do not exclude the possibility that a hypercholesterolemia might have existed for some time previously, and that the damage had already been done. The deposition of cholesterol and cholesterol esters, once it has occurred in the aorta of the adult person, is not reversible (Aschoff <sup>4</sup>).

Another possibility is that lipemia aids in producing a steatosis, as has been shown experimentally (Versé <sup>a</sup>). A relatively low cholesterol content may be more effective if a lipemia is associated.

The likelihood that in the young colored persons a hypercholesterolemia may have accounted for the greater inclination to atherosclerosis has already been discussed.

In the following diseases the fat angles are given for the entire group studied because of the lack of material. The average values given were so calculated as to correspond with the number of cases compared.

(b) Diabetes Mellitus: In diabetes mellitus the blood cholesterol and also the neutral fats are usually high (Klemper and Umbar, quoted by Bürger), especially when atherosclerosis of the aorta is associated (Joslin, Weiss and Minot). Klemper and Umbar, as well as Bürger, found that an acidosis was usually associated in cases of lipoproteinemia, although not always. Normal values of blood lipoid and cholesterol may be found in diabetes, especially when acidosis is not present. The low cholesterol values obtained by Hunt in diabetes mellitus with atherosclerosis of the aorta may be explained by the absence of acidosis at the time of examination, but they do not exclude the possibility that a hyperlipoproteinemia existed at some previous time when acidosis was present.

The finding of a hyperlipoproteinemia does not necessarily mean that an atherosclerosis of the aorta will follow, unless it is protracted over a long period. Joslin reports an increase of atherosclerosis in diabetes mellitus of from 30 to 68 per cent because the length of life has been increased from five to thirty years owing to the event of insulin. Whether it is the hypercholesterolemia or whether it is an alteration of the physical and chemical properties of the cholesterol ester resulting

from the disturbance of the cholesterol metabolism that influences the development of atherosclerosis of the aorta is not known.

The 8 persons examined for fat in the aorta were grouped together because of the lack of material (6 white and 2 colored persons).

Between 31 and 40 years there was 1 person with a fat angle of  $53.4^\circ$  as compared with  $44.3^\circ$  of the composite group (table 16). Between 41 and 50 years the F.A.A. was  $66.9^\circ$  as compared with  $48.2^\circ$ , and between 51 and 65 years it was  $59^\circ$  as compared with  $54^\circ$ . The average F.A.A. for diabetes mellitus was  $59.7^\circ$ , while for the composite group it was  $48.8^\circ$ .

In other words, the inclination to atherosclerosis of the aorta was much greater in those with diabetes mellitus than in the average group.

TABLE 16.—*The F.A.A. in Diabetes Mellitus*

Age	Cases	Fat, Gm.	F. A. A., Degrees	Average F. A. A.
31-40.....	1	0.288	53.4	44.3
41-50.....	3	0.720	66.9	48.2
51-65.....	4	0.751	59.0	54.0
		Average.....	59.7	48.8

TABLE 17.—*The F.A.A. in Chronic Glomerular Nephritis*

Age	Cases	Fat, Gm.	F. A. A., Degrees	Average F. A. A.
25-30.....	2	0.127	40.6	29.5
31-40.....	2	0.317	56.0	47.4
41-50.....	5	0.540	60.0	51.0
51-60.....	3	0.701	59.8	49.6
61-70.....				
71-.....	1	1.366	63.3	60.5
		Average.....	55.9	47.6

As the F.A.A. was  $59.7^\circ$ , this would indicate that the average diabetic patient examined at the Cook County Hospital has a moderate atherosclerosis. Unfortunately, the small number of cases makes the possibility of error very great. Yet the values given are far beyond the zone of error.

(c) Chronic Glomerular Nephritis: Bürger found high normal values of blood cholesterol in chronic glomerular nephritis, (200 mg. per hundred cubic centimeters of blood) using the Windaus digitonin method for the determination of these values. Other authors using a similar method have reported high normal or definitely increased values.

Thirteen persons with chronic glomerular nephritis were examined (5 white and 8 colored persons). The F.A.A.s are given in table 17. In comparing these results with those for the composite group it was noted that for every age the fat angle was higher in nephritis. The

average F.A.A. for nephritis was  $55.9^{\circ}$ , while for the composite group it was  $47.6^{\circ}$ , a 20 per cent difference.

Considering that the blood cholesterol was unchanged or only slightly elevated, the 20 per cent above the average inclination indicated that some other factor must have been present to account for this difference. The latter factor lies in the blood pressure and will be discussed in a later chapter.

(d) Chronic Tuberculosis: In chronic tuberculosis the cholesterol metabolism may be affected in many ways. When inanition sets in, a hypercholesterolemia may be present, which may then be accelerated by a cytolytic factor due to destruction of tissue. With extreme inanition the exhaustion of fat as well as of cholesterol may lead to a hypcholesterolemia, especially when a severe anemia is associated (Rosenthal and Patrzek).

The 25 persons with chronic tuberculosis (in 19 of whom the condition was pulmonary) were grouped together (8 white and 17 colored).

TABLE 18.—*The F.A.A. in Chronic Tuberculosis*

Age	Cases	Fat, Gm.	F. A. A., Degrees	Average F. A. A.	Weight, Lbs.
25-30.....	8	0.063	19.2	33.0	94
31-40.....	5	0.167	36.4	41.0	113
41-50.....	2	0.276	40.5	49.0	121
51-60.....	5	0.550	53.0	49.0	91
61-70.....	5	0.807	57.7	58.4	96
Average....			41.3	46.1	

Except for the first age group (from 25 to 30 years), the average F.A.A. did not vary greatly (table 18). Although inanition was present in most of the cases, it was not so marked that a hunger edema had set in. The average body weights given in table 5 demonstrate this point. Thus one might infer that there was some disturbance in the cholesterol metabolism.

The average inclination to atherosclerosis in persons with chronic tuberculosis was slightly lower ( $41.3^{\circ}$ ) than that of the average group ( $46.1^{\circ}$ ). A slight disturbance of the cholesterol metabolism, then, does not lead to atherosclerosis of the aorta, other factors being necessary.

(e) Carcinoma: In carcinoma, as in tuberculosis, the hypercholesterolemia is supposedly due to the lipemia from inanition and cellular destruction. Because carcinoma cells contain a high percentage of cholesterol (Wilheim and Fuchs), necrosis of the latter with absorption should lead to an even higher cholesterol content of the blood than in tuberculosis.

Among the 81 persons examined there were 61 white and 14 colored persons. Naturally the results for the white will be more accurate than those for the colored persons.

A slight difference was noted in the fat angles of the aorta (F.A.A.) for the various groups of white persons and for the entire group as compared with the average. The inclination to atherosclerosis was lower for the carcinoma group (table 19).

For the colored race the F.A.A. was much lower after the age of 30 than the average. Considering that a disturbance of the cholesterol metabolism was present, one would expect a greater inclination to atherosclerosis of the aorta. Here, as with tuberculosis, the blood pressure played a rôle, and it is evident that a slight disturbance of the cholesterol metabolism alone will not lead to a higher incidence of atherosclerosis of the aorta.

*3. Retention Hypercholesterolemia.*—Every condition that prevents the bile from entering the intestine leads to a retention hypercholesterolemia. In retention of long standing due to obstruction of the bile duct the blood cholesterol returns to normal (Stepp, Rosenthal and Holzer). The

TABLE 19.—*The F.A.A. in Carcinoma*

Age	White						Colored					
	Cases	Fat, Gm.	F. A. A., Average Degrees	Weight, Lbs.	F. A. A.	Cases	Fat, Gm.	F. A. A., Average Degrees	Weight, Lbs.	F. A. A.	Lbs.	
21-30												
31-40	7	0.141	31.0	34.0	126	3	0.167	49.5	40.7	88		
41-50	17	0.259	38.5	48.4	110	5	0.081	17.0	45.0	100		
51-60	22	0.324	37.3	51.0	134	3	0.273	40.2	44.0	101		
61-70	14	0.804	57.5	58.4	97							
71-	7	0.768	51.3	61.4	115							
		Average	43.1	50.1					Average	34.5	45.1	

hypercholesterolemia thus obtained extends over a short period of time and does not come under consideration in this paper.

(a) Atrophic Cirrhosis of the Liver: In atrophic cirrhosis of the liver without icterus an elevation of the blood cholesterol can also take place. The decreased amount of bile in the intestine plus the decrease in pancreatic lipase (associated cirrhosis of the pancreas) leads to a decreased absorption of cholesterol and fat, as has been shown by fat meal tests (Bürger and Habs). The blood cholesterol in atrophic cirrhosis of the liver may at first be elevated but returns to normal.

Of 7 cases of atrophic cirrhosis of the liver studied by Bürger and Habs, only 1 showed a blood cholesterol above normal, 299 mg. per hundred cubic centimeters, while the lowest value was 76 mg.

The 10 cases of atrophic cirrhosis of the liver without icterus that were studied (table 20) showed an inclination to atherosclerosis lower than the average, except between 51 and 60 years. The inclination for those with cirrhosis of the liver was about 22 per cent lower. Again the limited number of cases prevents definite conclusions, but there is a suggestion that when the cholesterol metabolism is slightly disturbed or not at all the inclination to atherosclerosis (F.A.A.) is not increased.

In atrophic cirrhosis of the liver with icterus there is a relative or an absolute decrease of the cholesterol esters in the blood (for the literature, Bürger and Thannhauser may be referred to). No such cases were available for study.

4. *Cytolytic Hypercholesterolemia*.—This type of hypercholesterolemia is supposedly dependent on cellular destruction. The absorption of the cholesterol-containing detritus should account for an increase of cholesterol in the blood.

In this group acute infectious diseases and lipoid nephrosis are considered. Tuberculosis and carcinoma, in which the hypercholesterolemia is of mixed types, have already been discussed. Some authors consider the hypercholesterolemia in the first two conditions mentioned as also of mixed types, i. e., transport and cytolytic (Bürger and Thannhauser<sup>b</sup>).

(a) Acute Infectious Diseases: Whatever the cause of the hypercholesterolemia may be, it is known that during the stage of fever the blood cholesterol is low, and that after the fever subsides the blood

TABLE 20.—*The F.A.A. in Atrophic Cirrhosis of the Liver Without Icterus*

Age	Cases	Fat, Gm.	F. A. A., Degrees	Average F. A. A.
29-40.....	2	0.079	17.5	35.0
41-50.....	2	0.213	32.8	48.5
51-61.....	6	0.571	53.2	50.5
Average.....			34.8	44.4

cholesterol rises. The more severe the infection the higher is the subsequent rise in the blood cholesterol (Grigaut and Bürger). This rise, according to the French school, is supposedly bound up with the immunologic reaction (Grigaut). Bürger and Thannhauser expressed the belief that a disturbance of the nutrition is the underlying factor.

As a rule the hypercholesterolemia is of short duration and should not affect appreciably the status of the aorta. In typhoid fever, however, in which it may extend over a period of a few months, fatty deposits in the aorta are common (Jores, Ophüls). In the 88 cases of acute infectious diseases examined there were no appreciable differences in the inclination to atherosclerosis (F.A.A.) as compared with the average for the white race (table 21). In the colored race there was a somewhat lower inclination between 25 and 30 years and 41 and 50 years. This difference, as will be explained later, was due to another factor present in the average group to a greater extent than in the group under consideration, viz., increased blood pressure.

From an experimental point of view the production of atherosclerosis by injections of killed bacteria has been reported by Saltykow and Klotz. A verification of this result could not be established by Starkodamski and Ssolowjew, who injected killed staphylococci and

added Klotz' technic of suspending the animal by its hindlegs daily. Inflammatory lesions were produced in the aorta after injections of bacteria by Thérèse and by Boinet and Romary. Benson, Smith and Semenov produced atherosclerosis of the coronary arteries of the heart in rabbits by injecting killed cultures of *Streptococcus viridans* only when cholesterol was added to the diet.

From an anatomic standpoint MacCallum found that persons dying of infectious diseases showed no special tendency to atherosclerosis.

The studies mentioned indicate that acute infectious diseases by themselves had no definite influence on atherosclerosis of the aorta, and when all other conditions were equal, the inclination to atherosclerosis was similar to that of the average group.

(b) Lipoid Nephrosis: Only 1 case of lipoid nephrosis was present in this series. It occurred in a colored man 55 years of age. The fat

TABLE 21.—*The F.A.A. in Acute Infections*

Age	White				Colored			
	Cases	Fat, Gm.	F. A. A., Degrees	Average F. A. A.	Cases	Fat, Gm.	F. A. A., Degrees	Average F. A. A.
25-30	4	0.047	12.0	19.6	7	0.076	24.3	43.0
31-40	16	0.185	39.6	34.0	4	0.229	46.2	44.2
41-50	13	0.308	46.3	48.4	10	0.182	28.4	46.0
51-60	8	0.422	45.2	51.2	8	0.373	41.4	48.2
61-70	9	0.553	46.8	54.4	3	0.786	56.9	62.0
71-	6	0.961	57.8	61.4				
	Average..		41.3	45.6	Average..		39.4	49.7

content of his aorta was 0.7784 Gm., which was interpreted as a severe atherosclerosis (ulcerations were also present).

The most important argument against the cholesterol metabolism theory in atherosclerosis of the aorta is that in the cases of hypercholesterolemia, notwithstanding the elevation of cholesterol in the blood from five to ten times above normal, atherosclerosis is not a common occurrence.

Several questions will have to be answered before this objection can be considered as valid.

According to Lawinowcz (quoted by Bürger), the diet does not affect the blood cholesterol in these cases. Bürger explained the hypercholesterolemia on the basis of a cellular destruction resulting from the activity of some infectious or toxic agent. It is to be seen whether or not the physical colloidal properties of the cholesterol derived from cellular destruction or other endogenous means are different from those of the exogenous cholesterol. Chalatow,<sup>a</sup> for example, has shown that in chemically similar cholesterol esters differences in their physical colloidal state may exist under similar temperatures.

The time element in many of these cases is too short, as the patients succumb early to the disease. If their lives could be prolonged, as in diabetes mellitus, e. g., it would then be seen whether the inclination to atherosclerosis also increases as has occurred in diabetes mellitus. Of course, the added factor of age would then be present, and it cannot be denied that senescence is of the utmost significance.

*Summary.*—A disturbed cholesterol metabolism as designated by hypercholesterolemia acting over a long period of time tends to increase the inclination to atherosclerosis of the aorta (diabetes mellitus, lipoid nephrosis). An increased inclination to atherosclerosis of the aorta may be present without a marked hypercholesterolemia (chronic glomerular nephritis). A slight increase in the blood cholesterol may not necessarily lead to an increased inclination to atherosclerosis, but possibly to a decreased inclination if the other influencing factors are wanting (tuberculosis, carcinoma, acute infectious disease).

*Comment.*—With a standard method of determining the inclination to atherosclerosis of the aorta (F.A.A.), a comparable method is needed for determining the cholesterol of the blood. Because of the various procedures in use for estimation of the blood cholesterol, the "normal" value shows variations from 85 to 310 mg. per hundred cubic centimeters of blood (Bang, Bloor methods). Such a marked variation speaks for a lack of uniformity either in the method employed or in the preparation of the patient before blood is taken for examination.

The recommendations given by Bürger for the preparation of the patient are given in detail because they are the best grounded.

The patient should not have eaten more than the average amount of fat and cholesterol (i. e., not over 100 Gm. of butter or over 3 eggs or their equivalent) the day before examination, and the last intake of fat should have been at least twelve hours before the blood is drawn. The serum thus obtained should be clear and transparent. The digitonin method as described by Windaus should be employed in the determination of the cholesterol.

There are no reports covering a large series of cases in which the aforementioned method was employed. That great fluctuations of the blood cholesterol according to age occur has been shown by György, who found in examining mother and child that the blood cholesterol of the child was about one-third that of its mother. An increase of the blood cholesterol after the first year has been demonstrated by Knauer. In adult life the variations are not so striking, but if differences occur, they are of great significance. Negative results would not, however, disprove the rôle of cholesterol in the production of atherosclerosis of the aorta, as it is possible that a disturbance of the cholesterol metabolism without an actual increase in the blood cholesterol may so alter the physical colloidal properties of the cholesterol (Chalatow<sup>a</sup>) that the end-result

would be similar or even greater. As will be discussed later, other factors enter in with age, such as the binding or precipitation of cholesterol in the aortic wall.

Only with such standard methods as those described can advances be made in the study of the rôle of the cholesterol metabolism in atherosclerosis.

In determining what part the diet plays in atherosclerosis it must be borne in mind that important factors are the amounts of cholesterol and of neutral fats consumed. Tables giving the percentage of cholesterol in various foods may be had in the work of McCollum and Simmonds, Thannhauser, and Bürger. The stress that the earlier authors placed on the protein content of the diet is not warranted, although proteins may elevate the blood cholesterol to some extent (Newburgh and Clarkson).

Subjects for study could be had in the various races who live on different types of food. More extensive studies could more readily be carried out with different religious sects who practice dietary restrictions. The monks studied by Saile are a good sample.

One attempt has been made to determine the blood cholesterol in natives of the Netherland Indies, whose diet is composed mainly of rice. De Langen found the blood cholesterol as well as the incidence of atherosclerosis to be low. Although this work was not carried out according to the standards indicated, it suggests that work in that direction may be fruitful.

Studies concerning endogenous cholesterol and atherosclerosis of the aorta can be more readily executed in large hospitals.

The plotting of the fat angles of the aorta as well as of the cholesterol angles of the blood for a large series of cases of each of various diseases would be exceedingly instructive. The values given in the text can be taken only as an indication of the possibilities, as the cases of many of the diseases examined were too few, and thus the results were subject to much error.

In conclusion, it appears that a disturbance of the cholesterol metabolism as determined by hypercholesterolemia, whether of endogenous or exogenous source, may alter the inclination to atherosclerosis.

*(To be continued)*

## SPECIFIC CHEMOTHERAPY FOR CANCER

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OMAHA

If malignancy is to be regarded as the result of fundamental metabolic abnormality through which cancer cells achieve their peculiar alterations of behavior, as seems probable, there is a possibility that the abnormality may be used as a basis for specific chemotherapeutic measures. Several of these have been suggested. Fischer-Wasels,<sup>1</sup> who was among the first to recognize this possibility, believed that it might be feasible to effect the cure of cancer by means of the intensive administration of oxygen. More recently, Roffo<sup>2</sup> attempted interference with the oxidoreductive mechanisms within the cancer cell, and Goldfeder<sup>3</sup> reported attempts at affecting the cancer cell by alteration of its hydrogen ion content.

A simpler line of attack would appear to be possible through the peculiarity of carbohydrate metabolism discovered by Warburg.<sup>4</sup> Either through the altered mode of utilization of dextrose in cancer tissue or through the altered affinity of such tissue for carbohydrate, it is conceivably possible to effect its selective intoxication by means of sugar compounds as carriers of toxic radicals. That some cancers at least show an increased affinity for carbohydrate was demonstrated in 1923 by Braunstein,<sup>5</sup> who observed that with the onset of cancer diabetic glycosuria frequently disappears. However, some results obtained in the course of the work reported here would apparently suggest that this relation is a variable one.

Theoretically, an ideal agent of this sort would be one in which the toxic radical is linked to one of the side-chains of the dextrose molecule. At present there does not appear to be any method by which a compound of this character can be prepared, and one is practically restricted to compounds in which acids derived from sugar are combined with a basic intoxicant radical. Also, the choice of the latter is somewhat limited, and in this work two only were used—lead and arsenic in basic form (tetramethylarsonium). While any of the arsonium compounds

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1. Fischer-Wasels, B.: *Klin. Wchnschr.* **7**:53, 106 and 153, 1928.
2. Roffo, A. H.: *Ztschr. f. Krebsforsch.* **37**:1, 1932.
3. Goldfeder, A.: *Ztschr. f. Krebsforsch.* **39**:421, 1933.
4. Warburg, O.: *Ueber dem Stoffwechsel der Tumoren*, Berlin, Julius Springer, 1926.
5. Braunstein, A.: *Deutsche med. Wchnschr.* **4**:880, 1923.

could be used in this manner, tetramethylarsonium was chosen because of its low general toxicity and, in part, because of its relative ease of preparation.

#### EXPERIMENTS

*Preparation of Agents.*—Tetramethylarsonium iodide was prepared by Cahours' method.<sup>6</sup> Arsenic trioxide and methyl iodide were heated in sealed bomb tubes to a temperature of from 165 to 175 C. for from twenty to twenty-four hours; after recovery of the unused methyl iodide by distillation, the residue was dissolved in strong caustic alkali, which was neutralized with concentrated hydrochloric acid. After allowing time for the separation of arsenic trioxide, this was filtered off, and the solution was evaporated to dryness on a water bath. From this product the arsonium halides were separated by successive solution in hot and freshly dehydrated ethyl and butyl alcohols. The residue left from evaporation of the latter solvent was dissolved in water and treated with an excess of silver oxide until the filtrate no longer reacted for halides, and the tetramethylarsonium hydroxide so obtained was purified by filtration and recrystallization.

Two acids derived from dextrose were used. Gluconic acid was obtained by the addition to a solution of pharmaceutic calcium gluconate of an equivalent quantity of oxalic acid, the precipitated calcium oxalate being removed by centrifugation. Qualitative tests showed traces of calcium but no oxalic acid. Glycuronic acid was prepared by the method of Quick<sup>7</sup> from the urine of dogs fed borneol. With both these acids and with one other the tetramethylarsonium salts were obtained by the addition to their solutions of the arsonium hydroxide to a point at which the solutions were neutral to litmus. None of these salts was obtained in crystalline form.

The lead compounds were prepared rather simply. The respective acids or their lactones were added to a suspension of an excess of lead carbonate, and the mixture was heated to boiling. The excess of undissolved carbonate was then removed by centrifugation. In addition to lead gluconate and glycuronate, salts were prepared from a number of miscellaneous sugar acids which were furnished through the cooperation of Dr. Fred Upson of the department of chemistry of the University of Nebraska. Of these, only one was tested in the form of the arsonium salt.

*Tumors.*—Transferable rat tumors, obtained for the most part through Dr. Francis Carter Wood of Crocker Institute, Columbia University, were used entirely for this work. In the earlier stages of the work studies were made on individual rats, but it developed quickly that, with respect to the agents which had any evident effect, the tumors came into two categories—slightly malignant ones, which yielded with some readiness to treatment, and others, more readily inoculable as a rule and more rapid in growth, with which results were obtained with more difficulty. Two representatives of this type were selected for the continuation of the work, which was then done with groups of rats handled similarly. These two tumors were the F R C carcinoma and the R39 rat sarcoma.

*Procedure.*—The agent, dissolved in physiologic solution of sodium chloride or, in later experiments, in distilled water, was injected intravenously through one of the caudal veins. The usual volume injected was 0.5 cc.; occasionally, with larger doses or in the case of relatively insoluble lead salts, 1 cc. was given.

6. Cahours, A.: Ann. d. chem. **122**:192, 1862.

7. Quick, A. J.: J. Biol. Chem. **74**:331, 1927.

*Lead Salts.*—Although work was first done on arsonium compounds, the results obtained with salts of lead are here presented first, in tabular form. As may be seen, with the exception of two or three of these, they are not particularly noteworthy; in spite of what would be in man a proportionately altogether excessive dosage, destruction of tumor was observed only occasionally, except with lead glycuronate, lead galactonate and, somewhat less strikingly, with lead glucoheptonate. On the basis of these results, tetramethylarsonium glycuronate and galactonate were used for further study. That the general lack of results with lead compounds may be due to precipitation of the lead in the serum is probable; however, test tube experiments indicated that such precipitation was slow, usually requiring forty-eight hours for completion.

*Tetramethylarsonium Salts.*—Gluconate: The initial test of this attempted method of therapy was made with tetramethylarsonium gluconate with a tumor derived in this laboratory, of slow growth and rather difficult transmissibility.

#### *Effects of Intravenously Injected Lead Salts on Implanted Rat Tumors*

Salts	Dose, Mg.	Tumor	No. of Rats	Size of Tumor	Results
Glucconate.....	5	FRC	4	From 3 to 8 mm. in diameter	Complete disappearance of 1
Glucconate.....	8	FRC	1	1.5 by 2 cm. in diameter	No effect
Gluconate.....	5	FRC	4	Palpable*	Complete disappearance of 2
Glucconate.....	5	R39	5	Palpable	Complete disappearance of 1
Glucconate.....	5	R39	4	From 1 to 3 mm. in diameter	Complete disappearance of 1
Glycuronate.....	5	FRC	4	Palpable*	Complete disappearance of all
Glycuronate.....	5	R39	4	Palpable*	Complete disappearance of 2
Arabonate.....	4.3	FRC	3	Palpable	Complete disappearance of 1
Arabonate.....	4.3	R39	4	Palpable*	Complete disappearance of 1
Galactonate.....	4.1	FRC	4	Palpable*	Complete disappearance of all
Galactonate.....	4.1	R39	4	Palpable*	Complete disappearance of 2
Glucoheptonate..	5	FRC	4	Palpable*	Complete disappearance of all
Glucoheptonate..	5	R39	4	Palpable*	Complete disappearance of 1
d-Mannonate....	5	FRC	4	Palpable*	No effect
d-Mannonate....	5	R39	3	Palpable*	Complete disappearance of 1
Rhamnohexonate	5	FRC	4	Palpable*	No effect
Rhamnohexonate	5	R39	4	Palpable*	No effect

\* These tumors were treated three days after implantation.

Only one of these tumors was available at the time of this test, but this was used, as no particular interest attached to its indefinite propagation, and there was no great expectation of a positive result. The tumor was 0.5 cm. in diameter. On the day following the injection of 5 mg. of the salt the tumor showed definite softening, and after palpation the animal became extremely intoxicated, with prostration and slow and shallow respiration—a degree of intoxication that was never observed subsequently, possibly because since then early palpation was avoided. Four days after the injection the tumor was represented by a boggy mass, and after eleven days it had completely disappeared. When the animal died of intercurrent disease about six months later the site of the tumor was represented by a small pigmented area of infiltrated leukocytes, principally mononuclear.

Three rats with tumors of strain J R S were given injections of 5 mg. of the arsonium salt; the tumors were 0.5 cm. in diameter. In two the tumors had completely disappeared within about three weeks; in the other there was temporary softening and shrinking, but later resumption and continuation of growth. Two rats with large tumors, 2 and 2.5 cm. in diameter, respectively, were similarly treated, without effect.

Only larger tumors of strain 256 were treated; these were from 1 to 2 cm. in diameter. Five milligrams was given to each of three rats, without effect. Two animals with tumors initially 1 and 1.5 cm. in diameter, respectively, received daily injections of 5 mg. of the arsonium gluconate for eight days; there was no apparent effect.

Eleven rats with tumors of strain F R C from 0.5 to 1 cm. in diameter were initially given single doses of 5 mg. of the tetramethylarsonium salt. In five the tumors had completely disappeared within three weeks. Of the others, one was again given an injection of the same dose after a four day interval, without effect. Another was given a second injection of 20 mg. after a three day interval. This animal died after six days, with progressive shrinkage of the tumor in the interim. Another animal which was similarly treated showed temporary softening and shrinking of the tumor, but later progression. One of the rats with a tumor initially 0.75 cm. in diameter received a daily injection of 5 mg. of the arsonium salt, beginning four days after the first injection. A total of nine injections was given. The tumor continued to increase in size, although when the animal was killed a month later it was almost wholly necrotic, measuring at this time 3 by 2 by 1.5 cm. Two animals were used for experiments with insulin, which will be described later. One other rat, with a tumor 1 cm. in diameter, was given a single injection of 30 mg. It died forty-five days later, with an almost wholly necrotic tumor measuring 1 by 1.5 by 2 cm.

Of ten rats in which tumors of strain R39 had been implanted three days previously and which received injections of 5 mg. of the arsonium gluconate, four showed complete disappearance of the tumors within three weeks. The others showed continued growth of the tumor.

Four rats with older tumors of delayed growth and of small size (from 1 to 5 mm. in diameter) showed no effects from a dose of 5 mg.

Two rats with larger tumors, 1.5 by 1.25 cm., which were given successive doses of 5 mg. and 20 mg. of the arsonium gluconate after intervals of two and four days, respectively, failed to show any effect on the tumors.

**Glycuronate:** With early tumors of strain F R C of three days' implantation disappearance of the palpable masses was observed in only three of ten animals after the injection of 5 mg. of tetramethylarsonium glycuronate. Larger tumors, from 0.5 to 1 cm. in diameter, which were treated by the injection of 10 mg., likewise reacted only occasionally to the treatment, progression taking place in all except two of the ten animals.

No effect was observable in four animals with three day tumors of strain R39 treated by the injection of 5 mg. of arsonium glycuronate. Another series of four animals with tumors 0.5 cm. in diameter received a dose of 10 mg., also without effect.

**Galactonate:** Eight rats with tumors of strain F R C, which had been implanted five days previously and had an average diameter of 0.5 cm., were given an injection of 5 mg. of tetramethylarsonium galactonate. All showed continued growth of the tumor.

Nine rats with tumors of strain R39 of three days' implantation received 5 mg. of the arsonium galactonate. The tumors were palpable in all the animals, and all showed progressive growth.

**Arsonium Salts and Insulin.**—In general, it must be stated that treatment of the more malignant tumors by arsonium salts of sugar acids failed to give particularly striking results. However, with the gluconate, disappearance of the tumors was observed with sufficient frequency and with a reaction so apparently immediate as to suggest an occasional selective affinity for the toxic agent—an

affinity that appeared to be more constantly present with some of the less malignant tumors used in the earlier work. Since it seems safe to assume that the agent after absorption should have approximately equal toxicity for cells of the several strains of tumor, it would appear that selective absorption is frequently lacking with the more malignant tumors. On the basis of the possibility that this inconstant lack of absorption was due to greater affinity of the tumor cells for carbohydrate, as a result of which this immediately available substance could serve to shield the tumor cells from its toxic modification, the next series of experiments was carried out in circumstances in which the available supply of carbohydrate was temporarily reduced by the simultaneous administration of insulin. Large but not intoxicating doses were used. The rat appears to tolerate insulin better than man, and in preliminary experiments it was found that 0.08 unit could be given to rats weighing slightly more than 100 Gm. without evidence of intoxication. In the course of the experiments this dose was occasionally given, and half this dose was frequently given, to rats weighing 80 Gm., without eliciting symptoms of hypoglycemia.

**Gluconate and Insulin:** Twelve rats with three day tumors of strain F R C were given injections of 5 mg. of tetramethylarsonium gluconate and 0.04 unit of insulin. In four there was evident softening of the tumors within four days and complete disappearance a week later. At that time the remaining rats, in which the tumors were now 0.5 cm. in diameter, were given 5 mg. of the arsonium salt alone. In five of these animals complete disappearance of the tumors followed; progressive growth occurred in three.

Six rats with tumors which had been implanted ten days previously and which at the time of injection were slightly less than 1 cm. in diameter, were treated with 5 mg. of the arsonium salt and 0.04 unit of insulin. Nine days later the tumors had disappeared in five. The remaining rat was then given an injection of 5 mg. of the arsonium salt alone. The second injection was without effect.

In an experiment conducted along with the foregoing, six rats with tumors which were similar in every respect were given an injection of 5 mg. of the arsonium salt. Within nine days complete disappearance of the tumors had occurred in four. The other two were then given an injection of 5 mg. of arsonium salt and 0.04 unit of insulin. Here, as in the preceding series, the second injection was without effect.

In two series of experiments, lots of twelve and ten rats with three day tumors of strain R39 were given an injection of 5 mg. of arsonium salt and 0.04 unit of insulin. In the first series, complete disappearance of the tumors occurred in all within about two weeks; in the second, complete disappearance occurred in all except one rat within the same time. The last animal showed progressive growth of the tumor.

Larger tumors failed to react to treatment: Twelve rats with tumors which had been implanted twelve days previously and which averaged about 1 cm. in diameter were given an injection of 20 mg. of arsonium salt and 0.08 unit of insulin. One of these rats died three days later, with definite evidence of hepatic intoxication. Continued growth of the tumors followed in ten rats, only one showing disappearance of the tumor.

Twelve rats with tumors which had been implanted seven days previously received 5 mg. of arsonium salt and 0.04 unit of insulin. Six of these animals showed definite growth of the tumors a week later; the other six showed doubtful growth. The animals were then given a second injection of 20 mg. of arsonium salt and 0.08 unit of insulin. There was disappearance of the tumors in four of these animals.

Glycuronate and Insulin: As arsonium glycuronate appeared less effective when used alone than the gluconate did, its combination with insulin was tried only with tumors of strain R39. Twelve rats with three day tumors were given 5 mg. of the arsonium salt and 0.04 unit of insulin. A week later none of the tumors showed softening, and most gave evidence of progressive growth. These animals were then given a second injection, half receiving 10 mg. of arsonium galactonate and half 10 mg. of the galactonate and 0.04 unit of insulin. The tumor continued to grow in all the rats except one, in which it disappeared.

Galactonate and Insulin: Eight rats with tumors of strain FRC, which had been implanted five days previously, were given an injection of 5 mg. of tetramethylarsonium galactonate and 0.04 unit of insulin. All showed progressive growth of the tumors.

Nine rats with tumors of strain R39, which had been implanted three days before, were given an injection of 5 mg. of the arsonium galactonate and 0.04 unit of insulin. There was disappearance of one tumor only; the others showed progressive growth.

Insulin Alone: Ten rats in which tumors of strain R39 had been implanted three days previously were given an injection of 0.04 unit of insulin. Uninterrupted growth of the tumors occurred in nine animals, with disappearance of the tumor in one.

#### COMMENT

The two tumors which were used for the greater part of this work must both be regarded as of rather high virulence. Of the two, strain R39 was decidedly of greater malignancy than strain FRC; it withstood implantation with practically complete success and was of very rapid growth, killing its host generally within about seven weeks. Strain FRC also withstood implantation well, but not with the uniform success observed with strain R39; it was of slower growth, and took considerably longer to cause death. These differences in behavior are reflected in the reactions of these tumors to the agents used in this study.

Several lead salts were found to cause complete disappearance of all the FRC tumors treated; with strain R39 the best achievement was the disappearance of 50 per cent of the small number of tumors studied. It was hoped that the response of these tumors would afford a guide in the selection of the best acid radical to be used with the tetramethylarsonium base. This was not the case, and results obtained with lead salts gave no clue as to the effectiveness of the arsonium compounds. It is possible that had the former been used in conjunction with insulin, an apparent relationship might have become manifest. But the best results obtained with the lead salts, with a relative dosage that would be utterly unsafe for a human being, lagged so far behind those obtained with arsonium gluconate that it was judged inexpedient to follow their investigation further.

Of the several arsonium salts studied, conspicuous success was obtained with one only—tetramethylarsonium gluconate. With this alone it was possible to cause the disappearance of relatively non-

malignant tumors in the small number of rats studied with some, but not absolute, regularity. With more malignant tumors this effect was occasionally manifest if the tumors were treated while still small. A much greater uniformity of success was obtained with these tumors, if small, when the arsonium salt was administered along with insulin. The fact that this procedure was more uniformly successful with the more malignant R39 tumor than with the F R C strain accords with the theory on which the coadministration of insulin is based. If the affinity of a cancer cell for carbohydrate is a function of its malignancy, or vice versa, with less malignant tumors less ability of the cancerous



Single tumor colony three days after implantation; numbers of these colonies appeared in the tissue encapsulating the embedded tumor mass, which was almost wholly necrotic.

tissue to avail itself of systemic reserve carbohydrate and so less chance of a shielding effect by this might be expected.

With strain R39, particularly, the only tumors treated with any uniformity of success were those which had been implanted a short time (three days) previously. Only rats with definitely palpable masses at this time were used. That actual implantation had occurred within this limited time is indicated by the photomicrograph, which shows one of a number of isolated tumor colonies in the peripheral tissue around an implanted fragment. The latter showed apparently complete necrosis. There has not been as yet an opportunity to determine the exact degree of progress that can be permitted these tumors

before successful treatment is no longer possible. As the results indicated, this is greater for strain F R C than for strain R39.

The toxicity of tetramethylarsonium gluconate for rats is low. While occasionally animals died comparatively shortly after receiving a 5 mg. dose, these deaths occurred so seldom that they must be regarded as due to intercurrent disease. The intoxication observed after this dose was acute and apparently entirely associated with disintegration of the tumor. Nor was early death observed after the fewer doses of 10 mg. A number of premature deaths followed a 20 mg. dose; the single animal that received 30 mg. died forty-five days later. When the gluconate was given in doses of 5 mg. on successive days, several animals received from 40 to 45 mg. without evidence of intoxication. Only one animal was observed with changes of tissue indicative of death from intoxication—a rat weighing about 80 Gm., which had received 20 mg. of the arsonium gluconate along with 0.08 unit of insulin; this was the only one of twelve animals of this series to succumb.

In the interests of brevity, no detailed account has been given of the course of the treated tumors. In general, disappearance of the tumor was preceded by a period of definite softening, with gradual absorption of the softened mass. At times there was direct shrinkage, with increased induration; recurrences were more likely to occur after this than after softening, though they occurred occasionally also with the latter. The relation of the disappearance of these tumors to the therapeutic procedure may perhaps be questioned, in view of the fact that spontaneous disappearance of implanted rat tumors is occasionally observed. In this work practically all the inoculated rats were subjected to some form of therapeutics, and the rate of natural disappearance, which is at best variable, but low, was not observed. Indirect evidence to the effect that treatment was responsible for the disappearance is furnished by the peculiar course of the latter—the rather prompt softening, followed by gradual absorption. Also, the relatively high rates of disappearance in the suitably treated rats were altogether disproportionate to those in animals treated otherwise.

There has been no opportunity to try the effects of combined arsonium salt and insulin therapy on native tumors. Several unsuccessful attempts were made to produce tar cancers in rabbits for this purpose, with tar obtained from the local gas-works. The tar appeared to be more toxic than cancerogenic, as all the animals died before or during the stage of benign papillomatosis. As to tumors in man, these would only rarely be suited to experimental trial. For this purpose it would appear necessary to select cases of early and superficially accessible recurrent malignant growth; late and otherwise hopeless cases would be useless, in view of the quantitative relation shown so clearly

with rat tumors. For the same reason, should the agent show any effectiveness with tumors in man, its use would appear to be as an adjunct to surgical removal of the great mass of the tumor tissue.

#### SUMMARY

On the basis of the theoretical possibility of effecting selective intoxication of cancerous tissue by means of carbohydrate compounds of toxic character, a study was made of the effects on transplantable rat tumors of various agents of this sort.

The toxic radical was represented by lead and tetramethylarsonium, the carbohydrate by a number of acids derived from hexose.

Results with the lead compounds were, in general, unsatisfactory; while it was possible to cause the disappearance of a number of implanted tumors of relatively low malignancy, with the most malignant tumor studied disappearance, on the whole, occurred only occasionally.

With the tetramethylarsonium compounds, the gluconate alone, when given in an apparently safe dose, caused the disappearance of a considerable proportion of a limited number of tumors of low virulence. With more malignant tumors, more effective results were obtained by the coadministration of insulin. When this treatment was used with the most malignant tumor studied, disappearance of the tumor followed in twenty-one of twenty-two cases.

If chemotherapy is to be effective, early treatment seems necessary. There appears to be a definite quantitative relationship between the effectiveness of treatment and the progress of the tumor, which has not been worked out in detail.

# THE RÔLE OF HISTAMINE IN INFLAMMATION

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The substance  $\beta$ -iminazolylethylamine, which is readily formed by the decarboxilation of histidine, has been given the name of histamine. This chemical material was first discovered by Yoshimura<sup>1</sup> in 1909, while in the following year Ackerman<sup>2</sup> produced it by allowing bacteria to act on the amino-acid histidine, and Barger and Dale<sup>3</sup> isolated it from ergot. In a recent communication, Best and McHenry<sup>4</sup> presented a detailed account of the many sources of this chemical body, and it is now concluded that histamine is a normal constituent of all body cells.

During the first few years following its discovery, the method of action and the effects of the drug were chiefly of physiologic interest. It was soon found, however, that there appeared to be a definite relationship between the effects of large doses of histamine, anaphylactic shock in animals and secondary wound shock in man. This discovery suggested the possibility that the amine in question, or some substance closely related to it, was either liberated or produced by the body under certain pathologic conditions. The work of Sir Thomas Lewis<sup>5</sup> definitely linked histamine to the study of physiology, pathology, immunology, dermatology and general medicine.

Cohnheim<sup>6</sup> was the first investigator to present the hypothesis that the vascular reaction is the factor of prime importance in an area of inflammation. In the monograph by Adami,<sup>7</sup> this question is discussed fully, but the process by which the phenomenon occurs is not explained. More recent work has suggested the possibility that the alteration in vascular structure and permeability which occurs at the site of an inflammatory lesion may be due to the effect of one or more of the products of cellular disintegration caused by the inflammatory agent. Since it is known that histamine is a normal constituent of all tissues and that it produces a vascular reaction similar to that observed in

\*J. J. Mackenzie Fellow in Pathology, University of Toronto.

1. Yoshimura, K.: Biochem. Ztschr. **28**:16, 1910.

2. Ackerman, D.: Ztschr. f. physiol. Chem. **65**:504, 1910.

3. Barger, G., and Dale, H. H.: J. Physiol. **40**:38, 1910.

4. Best, C. H., and McHenry, E. W.: Physiol. Rev. **11**:371, 1931.

5. Lewis, Thomas: The Blood Vessels of the Human Skin and Their Responses, London, Shaw & Sons, 1927.

6. Cohnheim, J.: Lectures on General Pathology, translated from second German edition by A. B. McKee, London, The New Sydenham Society, 1889.

7. Adami, J. G.: Inflammation, London, Macmillan and Company, Ltd., 1909.

inflammation, it has been suggested that this reaction is secondary to the liberation of the amine, irrespective of the type of irritant causing the inflammation.

Eppinger<sup>8</sup> described the reaction of the vessels of the skin to histamine as consisting of a threefold response: (*a*) a primary and local dilatation of the minute vessels of the skin, (*b*) a widespread dilatation of the neighboring strong arterioles, brought about entirely through a local nervous reflex, and, locally, (*c*) increased permeability of the walls of the vessels.

In his monograph, Lewis<sup>9</sup> demonstrated that the same type of inflammatory reaction can be produced by mechanical, electrical, thermal and other stimuli and concluded that the vascular changes are due to the effects of histamine or an H-substance liberated from the cells so stimulated. Dale<sup>10</sup> has drawn attention to the fact that the locus of the action of the amine on the vascular system shifts to a more proximal point as the biologic tree is ascended. The effects of histamine on the vessels of the dog, monkey and man have been studied by Burn and Dale<sup>11</sup> and others, who have concluded that in these species the amine produces a capillary and arteriolar dilatation and a constriction of the arteries and veins. Oertel,<sup>12</sup> in a recent publication, agreed with Ricker<sup>13</sup> that inflammatory hyperemia does not occur through a greater flux of blood and stated that this congestion is secondary to an arterial constriction. Though Oertel did not mention histamine, he appeared to be describing the same vascular state as that produced by this drug.

The early phenomena which occur in the inflammatory process, irrespective of the type of irritant, and which are dependent on the vascular reaction of the tissue, consist of: (*a*) dilatation and engorgement of the minute vessels, (*b*) exudation of fluid, (*c*) diapedesis of leukocytes and erythrocytes and (*d*) the formation of fibrin and an attempt to wall off the inflammatory focus. It therefore follows that if histamine can incite a true inflammatory response, it will reproduce all of the foregoing phenomena.

Eppinger, Lewis and others proved that the application of the amine to the cutaneous vascular system of dog and man results in a dilatation of the capillaries and arterioles and an exudation of fluid. Bloom,<sup>14</sup> Paul,<sup>15</sup> and Grant and Wood<sup>16</sup> reported that they were unable to show

8. Eppinger, H.: Wien. med. Wchnschr. **43**:1414, 1913.

9. Lewis,<sup>5</sup> p. 235.

10. Dale, H. H.: Lancet **1**:1179, 1233 and 1285, 1929.

11. Burn, J. H., and Dale, H. H.: J. Physiol. **61**:185, 1926.

12. Oertel, H.: Canad. M. A. J. **29**:378, 1933.

13. Ricker, G.: Frankfurt. Ztschr. f. Path. **33**:45, 1926.

14. Bloom, W.: Bull. Johns Hopkins Hosp. **33**:185, 1922.

15. Paul, J. R.: Bull. Johns Hopkins Hosp. **32**:20, 1921.

16. Grant, R. R., and Wood, J. E.: J. Path. & Bact. **31**:1, 1928.

that histamine was positively chemotactic. Wolf,<sup>17</sup> on the other hand, stated that the amine is strongly chemotactic in vivo and in vitro, and Weiss and his co-workers<sup>18</sup> produced leukocytosis by continuous intravenous injections of histamine in man. The question of the formation of fibrin about an area of reaction produced by histamine has received little or no notice. The recent work of Menkin<sup>19</sup> demonstrated that irrespective of the etiologic factor of its production a network of fibrin, occluding the intercellular spaces and the lymphatic channels, is formed about an inflammatory focus. On the suggestion of Professor Klotz, experiments were performed in an attempt to determine whether histamine is an agent calling forth all the characteristics of the inflammatory reaction and the manner in which it brings about its effect.

#### MATERIALS AND METHODS

All solutions used in the experiments consisted of histamine acid phosphate in physiologic solution of sodium chloride. Since solutions of histamine rapidly lose strength unless sterile (Best and McHenry<sup>4</sup>), all solutions were put in rubber-capped vaccine bottles and sterilized in the water bath. Tests for sterility and potency were made frequently.

In many of the experiments the capillaries of the skin were examined microscopically, and it was found that though only the minute, superficial vessels could be seen, with practice it was possible to observe accurately the changes which occurred in these structures. The skin was illuminated by a high power lamp, its rays being concentrated by lenses into a brilliant point of light. This light was filtered through a water cell containing a few drops of methylene blue. The filter, which cut out the heat rays from one end of the spectrum and the ultra-violet rays from the other, served to render the vascular structures more distinct as well as to protect the skin from the heat. A freely mobile, binocular, dissecting microscope was used. When practical, a drop of cedar oil was placed on the area to be examined, with a resultant marked increase in the clarity of the structures.

During the course of the experiments every care was taken to insure sterility of solutions, instruments and skin, so that the results obtained would not be complicated by infection. Very fine hypodermic needles were used and carefully inserted into the tissues in order to reduce the factor of trauma to a minimum.

#### EXPERIMENTS

In this experimental work the guinea-pig, rabbit, dog and human being were used. Since the results obtained in the guinea-pig corresponded to those in the rabbit, only the latter will be described. The effects of histamine on the vascular structures of the latter species are minimal. It was possible to cause a moderate dilatation of the conjunctival vessels by direct application of the amine, but injection failed

17. Wolf, E. P.: *J. Exper. Med.* **34**:375, 1921; *ibid.* **37**:511, 1923.

18. Weiss, S.; Robb, G. P., and Ellis, L. B.: *Arch. Int. Med.* **49**:360, 1932.

19. Menkin, V.: *Arch. Path.* **12**:802, 1931; *Arch. Int. Med.* **48**:249, 1931;

*J. Exper. Med.* **56**:157, 1932; *Proc. Soc. Exper. Biol. & Med.* **30**: 1069, 1933;  
*J. Exper. Med.* **57**:977, 1933.

to produce a wheal or a flare of the skin. In this work an attempt was made to produce an inflammatory reaction in the rabbit by applying histamine by various methods. The conclusion was reached that the amine, when repeatedly instilled into the conjunctival sac, applied to the intact, burned, scratched or cut skin for as long as seventy-two hours or repeatedly injected into the muscle of the rabbit, does not produce an inflammatory reaction.

The effect of histamine on human capillaries was investigated by performing the following experiment: A pneumatic cuff was placed about the arm and quickly pumped to above systolic pressure. This procedure rapidly produced a condition of circulatory stasis, and the microscopic appearance of the capillaries of the skin of the forearm was noted. A minute amount of histamine was injected intradermally, and there quickly appeared a small purplish spot at the site of the injection. Microscopic examination of this area showed an increase in number, size and tortuosity of the capillaries. This purplish spot did not spread so long as the pressure was maintained, but immediately after the pressure was released the spot extended rapidly and appeared as an irregular mulberry-colored area measuring about 1 cm. in diameter. This area showed a marked increase in number, size and tortuosity of the capillaries, with a sluggish flow of blood. The discoloration was transient, being rapidly obscured by the formation of a wheal which occurred at the site of the vascular dilatation. The wheal reached its maximum size in about five minutes, and the fluid was then gradually reabsorbed, so that it had entirely disappeared at the end of one hour.

The observations recorded suggest the following conclusions: The action of histamine on the minute vessels of the human skin produces a marked increase in number, size and tortuosity of the visible capillaries, with engorgement and a sluggish flow of blood through these structures. The release of pressure allows the amine to infiltrate the adjoining tissues and results in similar vascular changes. The formation of a wheal occurs at the site of vascular dilatation and is a result of serum exudation associated with the capillary damage caused by the amine. Ebbecke<sup>20</sup> showed that histamine produces a marked increase in permeability of the capillary wall during the period of wheal formation (about five minutes), and that at the end of this time the transudation ceases and a slow absorption of fluid commences. In his discussion on wheals, Lewis<sup>21</sup> stated that:

The outpouring of fluid into the tissue spaces is not the result of an increased filtration pressure. The increased permeability is not the result of simple stretching of the vessel's wall; it is the result of an independent change in the wall in response to stimulation, whereby this wall becomes unusually pervious.

20. Ebbecke, U.: *Klin. Wchnschr.* **2**:1725, 1923.

21. Lewis,<sup>5</sup> p. 80.

By comparing a site of the injection of histamine and an area of known inflammation, it is seen that the vascular structures of the two are similar in appearance. In each of these regions there is an increase in the number and tortuosity of the visible capillaries. The vessels are markedly engorged; the flow of blood through them is slow, and, in addition, both areas show an increased permeability for fluid.

The next experiment was performed in an effort to determine the effect of histamine on the migration of leukocytes, and was carried out as follows: The back of a dog was shaved, and into each of four well separated areas 0.1 cc. of a solution of histamine (1:500) was injected. One area was widely excised at the end of five minutes, another in ten minutes, the third at the end of one hour and the fourth at the conclusion of eighteen hours. The tissues were immediately placed in Orth's solution,<sup>22</sup> but the wheal which was present in the first two sections could not be preserved. Sections of tissue from the block removed five minutes after the injection of the amine showed many dilated capillaries, which were well filled with blood cells. The collagen fibers of the subcutaneous tissue presented no notable change, though in some areas they were somewhat separated. It was difficult to recognize the pathway of the needle through the tissues, since it was represented by only a small area of torn and distorted collagen fibers. There were no leukocytes in the tissue, and no diapedesis of erythrocytes had occurred. Sections of tissue removed ten minutes after the injection of histamine presented exactly the same changes as noted previously. The third block of tissue was also similar to the first two, except that the visible capillaries were not so numerous and were not engorged and distended with blood cells.

In the section of skin removed at the end of eighteen hours there was a definite inflammatory response. The track of the needle was represented by a small area of degenerating collagenous material, in and about which were scattered polymorphonuclear leukocytes and lymphocytes. This inflammatory cell infiltration was, however, not limited to the tissue immediately surrounding the pathway of the needle but extended well out to the periphery of the section. The number of leukocytes in any one field was found to decrease as sections more distal from the point of injection were examined. The vascular response was moderate in degree, and no diapedesis of erythrocytes had occurred.

The results of this simple experiment presented some interesting points. It was noted that one hour after the injection of histamine into the skin, at which time the effects of the amine were almost entirely dissipated, there was no cellular response. Though histamine had produced a vascular dilatation and exudation of fluid, at the end of one

22. Orth's solution consists of: bichromate of potassium, from 2 to 2.5 Gm.: water, 100 cc., and formaldehyde (40 per cent solution), 10 cc.

hour the sections of tissue presented no evidence to substantiate a hypothesis that the amine had a positively chemotactic force. Histamine infiltrates the tissues through the tissue and lymphatic spaces and produces the foregoing vascular reaction in all capillaries with which it comes in direct contact. The wheal begins at the point of injection and quickly follows the capillary dilatation. Subsequently, the fluid exudate, combined with the drainage of the amine through the blood and lymph stream, decreases the concentration of histamine in the tissues to a point at which it is no longer able to produce its vascular reaction. A fairly large dose of a strong solution of the drug was used, and it is apparent that the tissues immediately about the point of injection were subjected to a greater concentration of the solution of histamine for a longer period of time than were the more distant tissues; also, that both the strength of the solution and the time of action decreased toward the periphery of the wheal. In addition, the injection of the solution had two direct effects, the first being traumatization of the cells along the pathway of the needle and the second the rapid distention of the region by the exudation of fluid, with its resultant tearing of tissue and cell destruction. The production of the wheal served to cause further damage to the tissues, though since its formation was more gradual the effects were probably of a lesser degree.

The presence of the cellular infiltration in the section of tissue removed at the end of eighteen hours indicates that there was present in the tissues some substance which was positively chemotactic. The following possible sources of this attraxin must be considered: (a) a chemotactic substance liberated from the cells injured by the needle, (b) the histamine solution, (c) a product of cellular disintegration resulting from the direct action of histamine on tissues or (d) a product of cell destruction caused by the edema of tissues.

Histamine was injected into the shaved skin of a dog, and sterile needle punctures were made into several areas, both in and outside the flare. At the end of eighteen hours sections of skin were removed. The tissue into which histamine was injected presented the microscopic picture previously described, whereas the areas into which sterile needle punctures were made showed only a few scattered leukocytes about the wound. This reaction was not influenced by the presence of a flare. The experiment proves, therefore, that the chemotactic action of the products of cellular destruction caused by needling is practically negligible.

The next possible source of the chemotactic substance to be considered was histamine itself. It has been noted that there was no diapedesis of leukocytes after histamine had been allowed to act on the tissues and vascular structures for one hour. During this period the amine had been undergoing dilution and absorption, so that at the conclusion of the hour the effects of the drug had almost entirely dis-

appeared. By removing the skin of sterile blebs, produced by burning, it was possible to examine directly the vessels of the skin and to observe the effect of the direct application of the amine to these structures. The denuded areas were flooded with solutions containing various amounts of histamine, and it was possible to secure the aforementioned effects of the drug on the visible capillaries. Though the vascular dilatation and engorgement were continued for two hours, it was not possible to demonstrate any migration of leukocytes from the vessels to the fluid.

From the results of the foregoing work the conclusion was reached that the chemotactic substance in question was a product of cellular destruction. The cause of this damage to the tissue was due either to the effect of histamine on the cells or to the trauma to the tissue caused by the production of the wheals. To investigate this question, a wheal was produced by the injection of histamine into the skin of a dog's back, and in another area sufficient physiologic solution of sodium chloride to produce a similar-sized swelling was slowly injected. There was one factor which complicated this work and which it was impossible to obviate, namely, the liberation of histamine from the cells damaged by the injection of saline solution. However, this amount of histamine was very small, since it did not cause either a further wheal at the site or a surrounding flare. Since there was a large amount of fluid already present, any histamine liberated was greatly diluted, and this dilution was sufficient to prevent the appearance of any appreciable reaction to histamine.

Microscopic examination of the sections failed to demonstrate any difference between the areas into which the saline solution was injected and the areas in which a wheal was produced by histamine. Both tissues showed a diffuse infiltration of leukocytes with a slight concentration of white cells about the point of injection. This work shows that at the end of eighteen hours the cellular migration into an area of traumatized tissue, as produced by an injection of saline, was equal in degree to that which occurred as the result of an intracutaneous injection of histamine. In both of these areas there was present one common factor, namely, a rapid increase of fluid in a localized area of cutaneous tissue. This fluid caused tearing and distortion of the cellular structures, with the liberation of the products of tissue disintegration.

All microscopic sections were carefully examined to determine the amount of fibrin present. It was found that there was only an occasional strand of fibrin about the site of the injection of histamine and that there was just as much of this material at the site of the injection of saline solution. Though the reactions were allowed to proceed for as long as thirty-six hours, the formation of a network of fibrin in the tissue or lymphatic spaces was not demonstrated. In view of Menkin's work on the subject of inflammation, this result is interpreted as show-

ing that the application of histamine does not play an important part in this phase of the inflammatory process. Although, as Lewis has shown, the wheal fluid consists largely of the constituents of blood plasma, it is evident that the other factors concerned in the formation of fibrin are not active.

#### COMMENT

The foregoing experiments were performed in an effort to determine the rôle played by histamine in the production of inflammatory reactions in the skin. The effect of the cellular destruction due to trauma by the needle may be practically disregarded, since the skin punctured by a fine sterile needle consistently shows only a few leukocytes about the injured tissues. Lewis and his co-workers showed that the vascular reaction about a needle prick is dependent on the liberation of histamine from the injured cells. Histamine is, however, only one of the products of cellular disintegration, and the fact that it produces certain vascular phenomena is not sufficient reason for assuming that it is also the substance which attracts leukocytes into the tissues. No leukocytes were noted in the tissues one hour after an injection of histamine, and it has been shown by others that at the end of this period there is little histamine in the area into which the injection was made. The direct application of the amine to the vessels of the skin failed to attract leukocytes from the blood vessels. The experiments cited seem to prove that histamine is not positively chemotactic. Since no evidence was obtained to substantiate the hypothesis that histamine is positively chemotactic, and since the cellular infiltration of tissues damaged by the injection of saline solution is equal to the number of white cells at the site of a wheal produced by the injection of histamine, it was concluded that this chemotactic substance is a product of cellular disintegration other than histamine. The failure of the histamine reaction in tissues to give rise to the formation of any appreciable amount of fibrin is important when the part played by the amine in the process of inflammation is considered.

#### CONCLUSIONS

Evidence has been presented to show that histamine, when acting on the minute vessels of the skin of dog and man, causes capillary dilatation and engorgement with a slowing of the blood stream and an exudation of fluid. Injury and destruction of tissue, irrespective of the etiologic factor, will allow the liberation of small quantities of the amine, resulting in this reaction. The rôle played by histamine in the process of inflammation is confined to its effect on the small vessels. Histamine does not call forth the cellular elements in the inflammatory exudate. Further work is required to determine the action of various other products of cellular disintegration.

# Case Reports, Laboratory Methods and Technical Notes

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## MULTIPLE MALIGNANCY WITH METASTASIZING CARCINOID OF ILEUM AND MILIARY TUBERCULOSIS

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AND

OLIVE GATES, M.D.

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While we have shown<sup>1</sup> that multiple malignant tumors are far from rare, cases of triple malignancy are relatively infrequent; three such cases were recorded in our recent review, together with one newly reported case. We now report another, which is further noteworthy because of the coexistence of active miliary tuberculosis, claimed by Pearl<sup>2</sup> to be excessively rare in association with malignant disease, although Wilson<sup>3</sup> proved that Pearl's thesis is erroneous.

In addition, one of the three malignant tumors in this case was a metastasizing carcinoid. Raiford<sup>4</sup> found that carcinoid tumors represented only 0.18 per cent of gastro-intestinal tumors in his large series and that only one fifth had metastasized. Gaspar<sup>5</sup> summarized a few cases of metastasizing carcinoids of the small intestine and emphasized their rarity.

### REPORT OF A CASE

*History.*—A. W., an unmarried white woman, aged 53, entered the Palmer Memorial Hospital in 1929 under the care of Dr. L. S. McKittrick, complaining of a painless abdominal mass of four months' duration. At operation, a malignant papillary adenocystoma of the left ovary was removed, together with peritoneal implants from the cecum, appendix and parietes, which microscopic examination showed to be metastases of the ovarian tumor. Following intensive high voltage roentgen therapy the patient recovered and remained fairly well for four years. In 1934 she reentered the hospital, under the care of Dr. R. H. Sweet, with a history of progressive weakness and cramplike abdominal pains of three weeks' duration. These symptoms were regarded as due to metastases of the previously removed tumor, and no active treatment was attempted. Five days after entry the patient died with evidence of chronic intestinal obstruction.

*Autopsy.*—The peritoneal cavity showed adhesions between the omentum and the anterior abdominal wall; the appendix was absent; the diaphragm reached the fourth rib on the right and the fifth rib on the left; the surfaces were slightly dull.

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From the Laboratory of Pathology, New England Deaconess Hospital.

1. Warren, S., and Gates, O.: Am. J. Cancer **16**:1358, 1932.

2. Pearl, R.: Am. J. Hyg. **9**:97, 1929.

3. Wilson, E. B.: Am. J. Cancer **16**:227, 1932.

4. Raiford, T. S.: Am. J. Cancer **18**:803, 1933.

5. Gaspar, I.: Am. J. Path. **6**:515, 1930.

There was no free fluid or exudate. The sigmoid colon was dilated so that it extended almost up to the left costal margin and measured approximately 10 cm. in diameter. At the upper end of the dilatation the colon was twisted on itself, occluding the intestine, but without any surrounding inflammatory or degenerative reaction. There was an unusually long mesentery for the descending colon and sigmoid, which was twisted clockwise for nearly a complete revolution. The serosa of the lower dilated sigmoid was markedly congested. No mechanical obstruction was found in the pelvis. A few scattered firm nodules averaging 1 cm. in diameter were present in the broad ligament. The lymph nodes were palpable in the mesentery of the small intestine. Six firm, gray polypoid nodules, varying from 1.5 to 0.3 cm. in diameter, apparently arose from the mucosa of the ileum and descending colon. The sigmoid colon had a smooth stretched out mucosa in the upper portion and a congested dark red hemorrhagic mucosa with irregular foci of erosion in the lower sigmoid colon and in the rectum. Several thrombosed submucosal vessels were prominent. Except where ulcerated, this involved mucosa had a rather glassy turgid appearance. The right lung weighed 545 Gm.; the left, 520 Gm. In the left upper and lower lobes were a few scattered nodules of firm, glistening, semitransparent tissue which slightly everted on section, the largest being 1 cm. in diameter. In addition similar nodules about 0.2 cm. in diameter were scattered throughout the lung, giving it a sandy texture. No large nodules were found in the right lung, but there were similar fine nodules scattered throughout all the lobes. The suprarenal glands were normal except for a glistening white, firm, fairly discrete nodule 0.7 cm. in diameter at the tip of the right gland involving the medulla and extending into the cortex. The right kidney weighed 380 Gm.; the left, 275 Gm. The surfaces appeared smooth and pale when the capsules were stripped. The cortices were 0.6 cm. wide. In the tips of the pyramids were yellowish-white, smooth, irregular foci, the largest of which was 0.5 cm. in diameter. The pelvis and ureters were normal. Involving the central portion of the right kidney was a cyst 5 cm. in diameter extending from the pelvis to 1.5 cm. beyond the surface of the kidney. It was filled with gelatinous yellow material. The parenchyma was slightly compressed around the cyst. The uterus was normal except for three very slightly raised, soft foci 0.6 cm. in diameter which extended 0.2 or 0.3 cm. below the surface. The bodies of the fourth and fifth lumbar vertebrae were white and contained foci of necrosis. White thick fluid raised the anterior periosteum; the cartilaginous disks were also invaded. The marrow of the upper lumbar vertebrae was dark red.

*Microscopic Examination.*—Scattered throughout both lungs were tubercles varying considerably in both size and age. The largest were caseous and had considerable surrounding fibrosis. Tubercles were found on the mucosal surface of many bronchi. In addition there was a focus of dense fibrous tissue with central calcification. Tubercles, chiefly early ones, were scattered throughout both the pulp and the corpuscles of the spleen. In the submucosa of the ileum there was a tumor nodule consisting of uniform, oval, medium-sized cells with no definite arrangement. These cells were not encapsulated and extended slightly into the mucosa and muscularis. Rare mitoses were present. Silver stain showed numerous argentaffin granules. In the colon the tubercles in the mucosa and submucosa did not show any definite ulceration. A polypoid mucosal nodule formed of closely packed, hypertrophied, atypical, mucosal glands invaded the muscularis. The cells showed mucous secretion, and mitoses were present. The sigmoid colon showed marked congestion and edema of all the coats with hemorrhage and necrosis of the mucosa. There was a diffuse infiltration of lymphocytes, plasma cells and polymorphonuclear leukocytes. Fibrin covered the serosal

surface. The liver was normal except for early tubercles in or near the majority of the portal spaces. The right suprarenal gland showed destruction of two thirds of the tissue by caseous necrosis with peripheral small tubercles, giant cells and fibrosis. Tubercles were scattered throughout the medulla of the kidney and a few scattered glomeruli were sclerotic. The uterus showed one small tubercle in the myometrium. The mesenteric lymph node was almost entirely replaced by tumor cells identical with those in the ileum. The hilar lymph nodes were replaced by dense fibrous tissue; there were a few scattered tubercles at the periphery. The bone marrow showed foci of caseous necrosis with peripheral giant cells and extensive destruction of the bone.

*Anatomic Diagnosis.*—The following diagnosis was made: resection of the ovaries for malignant papillary adenocystoma with peritoneal metastases; multiple carcinoid tumors of the small intestine with metastasis to the mesenteric lymph nodes; malignant adenoma of the colon; miliary tuberculosis of the lungs, spleen, suprarenal glands, kidney, myometrium and liver with tuberculosis of the colon, vertebrae and lymph nodes; torsion of the sigmoid colon with necrosis and hemorrhage; thrombosis of the superior hemorrhoidal artery; retention cyst of the right kidney; cholelithiasis and arteriosclerosis.

## General Review

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### THE PROBLEM OF INTRANUCLEAR INCLUSIONS IN VIRUS DISEASES

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ST. LOUIS

In trying to present this problem the German method of *Fragestellung* is used. The questions do not reveal information so much as they do the lack of it.

#### *What are viruses?*

The term "virus" is simply the Latin word for poison. Virus diseases are caused by substances whose organismal nature has not been proved. As soon as any virus is found to be a micro-organism it is no longer considered a member of the group. Some imagine viruses to be living agents, like those with which one is familiar, but smaller and more elusive. Others think they behave more like inanimate substances, such as enzymes. Still others regard viruses as living things of a type altogether new to present understanding. Be this as it may, viruses certainly occur in thousands in plants as well as animals. In attempting to learn something about them by the cellular responses which they provoke, my colleagues and I have concentrated attention on those which lead to the production of intranuclear inclusions.

#### *What are inclusions?*

Cytologists and virologists employ the word "inclusion" differently. By the former it is commonly applied to nuclear and cytoplasmic bodies that have nothing to do with viruses. According to Maximow and Bloom,<sup>1</sup> cytoplasmic inclusions comprise: "accumulations of proteins, fats, carbohydrates, pigments, secretory granules, chromophile substance and crystals." They are often looked on as "passive, lifeless, temporary constituents of the cell." Nuclear inclusions are of the same order. Reference may be made to fat droplets, crystals and melanin.

Virologists have simply appropriated the word "inclusion." Their original idea was that the inclusion material was of extracellular origin

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From the Anatomical Laboratory, Washington University;

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Presented as the first Annual Harry Hayward Charlton Lecture in Anatomy, University of Missouri, May 21, 1934.

1. Maximow, A., and Bloom, W.: Textbook of Histology, Philadelphia, W. B. Saunders Company, 1931.

—a view not borne out by recent work. In order to avoid ambiguity some now refer to the inclusions in question as "virus inclusions," which is satisfactory provided that it does not carry the impression that the inclusions are made of virus. It is more correct to speak of "inclusions in virus diseases." To do so, however, is not practicable because the phrase is cumbersome. In characterizing nuclear inclusions it is customary to emphasize: (1) the acidophilic staining of the inclusion mass, (2) the presence of a clear halo between it and the nuclear membrane and (3) the margination of basophilic chromatin on the nuclear membrane.

*In what conditions are inclusions found?*

It would be tedious to list each and every condition separately. There is, however, a range from severe epidemic diseases, like yellow fever, smallpox and many others, to mild infections called "inapparent" because their presence is not revealed by noticeable clinical symptoms, as the salivary gland disease of guinea-pigs.

Nuclear inclusions have been observed almost by chance in occasional individuals of all species whose tissues have been repeatedly examined: man, monkeys, dogs, guinea-pigs, rats and mice. Whether in each case their presence indicates virus action is uncertain. Since the occurrence of nuclear inclusions without signs of disease is just as important, in a statement of the whole problem, as their presence in disease so devastating that they irresistibly attract attention, an attempt has been made to secure data bearing on their distribution in wild animals also.

E. J. and L. E. Rector<sup>2</sup> discovered that all of fourteen common garden moles collected near St. Louis exhibited nuclear inclusions in their salivary glands. These proved to resemble closely the well known inclusions in guinea-pigs, young children and rats. There is every reason to suppose that further investigations will bring to light similar modifications in other species, likewise in the absence of revealing clinical symptoms.

I decided to go further, in cooperation with the Philadelphia Zoological Society. Dr. Fox, the pathologist in charge, Dr. Lucas and I have just completed a survey of many mammals and birds with special reference to the kidneys.<sup>3</sup> Nuclear inclusions something like those caused by the virus of yellow fever were seen in the kidneys of several parrot-like birds of Central America, termed "Guatemalan

2. Rector, Eleanor J., and Rector, L. E.: Intranuclear Inclusions in the Salivary Glands of Moles, Am. J. Path. 10:629, 1934.

3. Fox, H.; Cowdry, E. V., and Lucas, A. M.: The Distribution of Intranuclear Inclusions Suggestive of Virus Action in Wild Animals, Am. J. Path., to be published.

amazons;" but they were never so frequent per unit volume of tissue as the yellow fever inclusions, nor were they accompanied by the same severe tissue reaction. Other inclusions, also belonging to type *A* (see following question), were found, while inclusions of type *B* were frequently encountered. That the latter are all due to virus action is questionable.

It is likely that nuclear inclusions are widely distributed in vertebrates with, but more often without, evidence of disease. The nuclear inclusions in polyhedral virus diseases of insects are interesting but atypical.

*Can inclusions be classified in groups?*

The reactivity of the nucleus is limited by the cytoplasmic buffer interposed between it and the environment and by the relative uniformity of the chemical substances of which nuclei are made and from which the reaction products must be built, unless outside materials enter and contribute to the formation of the inclusions, for which there is no evidence except in regard to water. Yet nuclear inclusions are not all alike by any means. Since it is risky to try to classify them on the basis of published descriptions, only those under investigation in this laboratory, or in preparations graciously sent to me by other workers, are considered.

Type *A* inclusions occur in:

1. Herpes
2. Yellow fever
3. Chickenpox
4. Virus III infection
5. Salivary gland disease of guinea-pigs
6. Salivary glands of moles
7. Kidneys of Guatemalan amazons
8. Fox encephalitis (from R. G. Green)
9. Whooping cough (from H. A. McCordock)
10. Salivary glands of rats (from Juanita Thompson)
11. Salivary glands of children (from S. B. Wolbach)
12. Infectious tracheitis of chickens (from Oskar Seifried)
13. A disease of parrots and parrakeets (from J. R. Meyer)
14. Kidneys of frogs (from Baldwin Lucké)
15. Brain of mouse with louping ill (from L. T. Webster)
16. Mad itch in dogs (from E. W. Hurst)
17. A disease of owls (from R. G. Green)
18. Many species in the absence of disease

The nuclear reaction is total and proceeds to complete degeneration. The inclusions are amorphous or particulate, but may be condensed in rounded masses. The ground substance of the entire nucleus is profoundly disturbed, and all of the basophilic chromatin eventually margi-

nates on the nuclear membrane, except in the case of the salivary gland inclusions in moles, which are more basophilic than acidophilic. After fixation the material of which the inclusions are constructed is not easily removed by acetic acid, alcohol, chloroform and other solvents. It contains little or no masked iron or thymonucleic acid.<sup>4</sup> Incineration shows that the yellow fever inclusions are devoid of mineral matter.<sup>5</sup> L. E. and E. J. Rector<sup>6</sup> have found that the same observation holds for mature herpetic inclusions. In interesting contrast, a large amount of mineral, especially calcium, occurs in the nucleoli and basophilic chromatin.

Type *B* inclusions occur in:

1. Poliomyelitis
2. Borna disease (from I. A. Galloway)
3. Kidneys of sewer rats (from E. Hindle)
4. Livers of mice (from G. M. Findlay)
5. Rift Valley fever (from J. R. Hudson)
6. Gliomas (from D. Russell)
7. Many species unaccompanied by evidence of disease

The reaction is localized in certain areas of the nucleus, where acidophilic droplets make their appearance. These often look hyaline and may be of small or large size. The nucleoplasm in which the inclusions are embedded may not be noticeably altered. Basophilic chromatin fails to marginate on the nuclear membrane. It may even accumulate to some extent on the centrally placed inclusions. The process seldom goes on to complete nuclear degeneration, and it is not accompanied by the marked reaction of tissue frequently but not always present with the type *A* inclusions. Such inclusions can be distinguished from nucleoli by: (1) recognition in the same nucleus of nucleoli stained differently; (2) their range of variation in number and size; (3) absence in them of detectable amounts of mineral and so on. It is unsafe to assume that different *B* inclusions are of similar composition. Their analysis has not been energetically pushed.

*Are inclusions of type *A* and *B* distinct or simply different expressions of the same process?*

That the latter may be the case is shown by preparations of rabbit encephalitis kindly submitted by Dr. C. C. Levaditi, which are exceptional in that they contain both. The material available seems to indicate that the type *B* inclusions represent less drastic modifications which may develop slowly and do not necessarily end in nuclear distintegration.

4. Cowdry, E. V.: Science **68**:40, 1928.

5. Cowdry, E. V.: Am. J. Path. **9**:149, 1933.

6. Rector, L. E., and Rector, E. J.: Am. J. Path. **9**:587, 1933.

In the animal series as a whole they are of much more frequent occurrence than the type *A* inclusions. Evidence that they are related to a virus, though satisfactory in some cases, is often lacking.

*How far do variations in histologic technic modify the appearance of the inclusions?*

No special technic is required for the identification of nuclear inclusions. They may be seen in sections of tissues fixed in formaldehyde solution or in Zenker's fluid and stained with hematoxylin and eosin or with eosin and methylene blue, indeed by any method which brings to light the so-called acidophilic and basophilic materials. Recognition depends on easily detectable morphologic alterations in the nucleus as much as it does on changes in staining reaction. Only the inclusions caused by the virus of yellow fever have been investigated in detail in the fresh state, though several of the others have been seen. Fixation does not much modify either the morphologic character or the arrangement of the inclusions. But some fixatives, like Zenker's fluid, produce an additional coagulation of the nucleoplasm not seen after the use of fixatives rich in osmic acid.<sup>7</sup> Obviously preservatives leading to a swelling or a shrinkage of the nuclei alter the apparent width of the clear halo about the inclusions. Quite noticeable is the influence of the fixative on the staining reaction of the inclusions. Ordinarily the inclusions are acidophilic. Following treatment with Carnoy's fluid (absolute alcohol 6; chloroform, 3; acetic acid, 1) they are often distinctly basophilic.<sup>8</sup> When the color contrast is sharp, the inclusion material is generally less intensely stained with acid dyes than the acidophilic nucleolus or plasmosome.

*Is there a sharp line of distinction between the formation of inclusions and nuclear changes unrelated to viruses?*

Acidophilic material normally occurs in a great many nuclei, especially in those of nerve cells. In ordinary oxychromatic degeneration this increases in amount, though the mode of increase seems to differ, in a manner hard to specify, from that involved in the development of type *A* inclusions. A clear halo is usual about the mature type *A* inclusion which is not seen about the acidophilic material in cases of oxychromatic degeneration. This difference may result from the fact that when nuclei are first affected by this group of viruses they increase in size slightly or very considerably—an alteration not found in oxychromatic degeneration. In later stages of the reaction they usually shrink, and the halo becomes obliterated, so that the resemblance to oxychromatic degeneration is quite striking. While margination of chromatin also takes place in oxychromatic degeneration it does so at a later time,

7. Cowdry, E. V., and Kitchen, S. F.: Am. J. Hyg. **11**:227, 1930.

8. Cowdry, E. V.: Arch. Path. **10**:23, 1930.

as a rule after the nuclei have begun to shrink; it is rare to find all the central parts of large, robust or hypertrophied nuclei swept clear of basophilic chromatin in quite the same fashion as in these virus diseases. Though a sharp distinction cannot be made, and it appears that one is dealing with a process of nuclear disintegration peculiar in only minor respects when brought about directly or indirectly by viruses, there are, nevertheless, some marked differences. In the formation of type *A* inclusions the nuclei are always successively involved. Some look altogether normal, others show early stages in the reaction, and still others, late stages; whereas in oxychromatic degeneration most nuclei belonging to neighboring cells of the same sort exhibit exactly the same changes.

In autolytic postmortem nuclear degeneration some acidophilic material may be seen, depending on the kind of nucleus involved, and the basophilic chromatin eventually fades away without much margination. This change also spreads over nuclei of a given type almost uniformly, which again differs from the individuality in the formation of type *A* inclusions.

Since the nuclear alterations that accompany the formation of type *B* inclusions are less marked, there are fewer related properties to contrast. The acidophilic bodies that my collaborators and I have seen in the nuclei of hepatic cells of 22 per cent of dogs examined look, at first sight, like type *B* inclusions because the chromatin is not typically marginated, simply being pushed aside, and complete nuclear degeneration does not follow; but they are distinctly crystalline (see Brandts' figures<sup>9</sup>). In the epididymis the nuclei undergo remarkable alterations not so regularly evident anywhere else in the body, which have been interpreted by some as indicating the formation of a secretion product, and by others, as being degenerative products. The nuclei in the epididymis illustrated by Hammar,<sup>10</sup> Heidenhain and Werner,<sup>11</sup> Benoit<sup>12</sup> and Ludford<sup>13</sup> contain masses of material that very closely resemble type *B* inclusions. Another region where nuclear inclusions apparently occur constantly in man and certain species of animals, in the absence of disease, is in nerve cells of the nucleus supra-opticus and paraventricularis of the midbrain. Details and a review of the literature, which extends back many years, may be found in the papers of Scharrer and Gaupp<sup>14</sup> and of Scharrer.<sup>15</sup> The inclusions are interpreted by the authors

9. Brandts, C. E.: Beitr. z. path. Anat. u. z. allg. Path. **45**:457, 1909.

10. Hammar, J. H.: Arch. f. Anat. u. Entwickelungsgesch. (supp.) **1**:42, 1897.

11. Heidenhain, M., and Werner, F.: Arch. f. Anat. **72**:556, 1924 (fig. 9).

12. Benoit, M. J.: Arch. d'anat., d'hist. et d'embryol. **5**:175, 1926 (fig. 80).

13. Ludford, R. J.: Proc. Roy. Soc., London, s. B **98**:353, 1925 (fig. 11).

14. Scharrer, E., and Gaupp, R.: Ztschr. f. d. ges. Neurol. u. Psychiat. **148**: 766, 1933 (fig. 2).

15. Scharrer, E.: Frankfurt. Ztschr. f. Path. **27**:135, 1934 (fig. 5).

as intranuclear stages in the formation of a secretory product, and the claim has been advanced that the area is to be regarded as constituting an additional endocrine organ, the *Zwischenhirndrüse*. The bodies in the epithelium of the stomach figured by Carlier<sup>16</sup> resemble Borna inclusions. If such inclusions were found regularly in a virus disease and were absent in normal controls they would certainly be classified as type *B* inclusions.

*Do all nuclear inclusions indicate virus action?*

Types *A* and *B* are of a different status. It was with reference to inclusions like those in the salivary glands of guinea-pigs and in herpes, which belong to type *A*, that Cole and Kuttner<sup>17</sup> expressed the view that when typical inclusions are found, the presence of a filtrable virus is to be assumed unless its absence can be proved experimentally. This is going too far, but one naturally looks for a virus when such inclusions are observed. Often a virus has been discovered, but there are exceptions. These failures will be interpreted differently depending on experience and point of view. Some will say that the conditions were not favorable for the demonstration of virus action, while others will entertain the possibility that some type *A* inclusions may be produced in the absence of a transmissible virus. The question of how this may conceivably be done will be considered later.

With inclusions of type *B* the probability of a virus etiology is rather less. While a few of them appear regularly in cells injured by certain viruses, the vast majority are not known to be associated with viruses. Wolf and Orton<sup>18</sup> observed nuclear inclusions resembling those in poliomyelitis in many other diseases of the nervous system. Such inclusions seem to crop up without rhyme or reason, as shown by the same authors' studies on brain tumors<sup>19</sup> and by our own survey of the tissues of wild animals. Consequently with these type *B* inclusions the existence of a virus should not be taken for granted. They may be simply the expression of nuclear modifications occurring not only in some virus diseases but also in many conditions for which viruses are probably not responsible.

*Does the same virus produce similar inclusions in cells of different types?*

As a rule viruses are rather selective in their action. The virus of herpes is one of the most "cosmopolitan." The inclusions which it produces in a wide variety of cells are similar and of type *A*. When the submaxillary virus of guinea-pigs is injected intracerebrally it leads

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16. Carlier, E. W.: Cellule **16**:405, 1900 (plate III).

17. Cole, R., and Kuttner, A. G.: J. Exper. Med. **44**:855, 1926.

18. Wolf, A., and Orton, S. T.: Bull. Neurol. Inst., New York **2**:194, 1932.

19. Wolf, A., and Orton, S. T.: Bull. Neurol. Inst., New York **3**:113, 1933.

to the formation of inclusions true to the original type *A* in the salivary glands, but without the extraordinary hypertrophy and the accompanying cytoplasmic inclusions which are marked features of the salivary gland reaction. No records are available as to the action of the viruses that produce type *B* inclusions on cells different from those primarily affected. The factor leading to their development is more local and does not exhibit the same spread to other tissues. As far as an answer is possible it is, therefore, in the affirmative.

*How dissimilar are inclusions produced by different viruses in the same cell types?*

If one excludes all other manifestations of virus action, are the inclusions of themselves pathognomonic? A definite answer is available only for those viruses which can be induced to act not simply on the same kinds of cells but also on individuals of the same species. This requirement has been fulfilled in only two instances. Herpes virus and virus III produce in the testicle of the rabbit inclusions of type *A* which are slightly different in degree of basophilia, while the herpetic virus and the virus of yellow fever, acting on the hepatic cells of the monkey (*Cebus*), cause the development of inclusions of different morphology. If identity of species is not insisted on, the material on which this paper is based permits a comparison of inclusions caused by a number of viruses in nerve cells, renal cells and hepatic cells; but the comparison is not so close as in the rabbit testicle, for the respective tissues were not fixed in the same bottle, embedded in the same block of paraffin and the sections mounted and stained on the same slide.

In nerve cells the inclusions produced by the Borna virus in horses, the virus of mad itch in dogs, the virus of louping ill in mice and the virus of poliomyelitis in monkeys are quite different. But to distinguish those in herpes, mad itch, the rabbit encephalitis of Levaditi and in guinea-pigs' brains inoculated with the salivary gland virus, which are all of type *A*, would not be so easy. It has not been tried under controlled conditions. In renal cells the inclusions caused by the Lucké virus in frogs and the Hindle virus in rats are not at all the same. In hepatic cells the inclusions excited by the herpetic and yellow fever viruses (already alluded to) and by the virus of fox encephalitis are different in shape and arrangement. To distinguish, however, the inclusions in owls and in parrots and parrakeets from the herpetic or yellow fever ones may be difficult or impossible. On the other hand, the inclusions occurring without obvious explanation in dogs<sup>20</sup> are so similar to those in fox encephalitis<sup>21</sup> as to suggest that the same virus may be responsible.

20. Cowdry, E. V., and Scott, Gordon, H.: Arch. Path. 9:1184, 1930.

21. Green, R. G.; Kalter, M. S.; Schillinger, J. E., and Hanson, K. B.: Am. J. Hyg. 18:462, 1933.

*What is the site of action of these viruses and what is their means of spread?*

In these, as in so many other respects, viruses behave differently. That some of them pass through the cell membrane and enter the cytoplasm is likely, but whether they always do so is one of the many unanswered questions. In late stages of the disease reaction, when the concentration of virus is greatly increased, and more and more cells become involved, the injury may be primarily due to the liberation of toxic substances which may operate in many ways. It is possible that they go one step farther and enter the nuclei. The original contention of Goodpasture and Teague<sup>22</sup> that the inclusions are masses of virus is, however, giving way.

Mechanical injury frequently opens the path for cytoplasmic invasion. Goodpasture<sup>23</sup> furnished interesting evidence that the herpetic virus may enter the traumatized peripheral processes of nerve cells and travel all the way to the nerve cell bodies, not in the tissue spaces between the nerve fibers, but in the substance of the axons or dendrites within the myelin sheaths. The physical factors involved are difficult to comprehend. The processes are tubes of capillary size through which no chemical substance could travel without being arrested by practically complete adsorption on the walls. Moreover, the microdissections of de Renyi<sup>24</sup> show that the axonic substance is of gelatinous consistency. When the myelin sheath is removed, it retains its filamentous shape and can even be cut in segments, which likewise hold their form. It is not an instance of passage through a thin aqueous material of low viscosity. One is obliged to hypothecate a spread by local increase in amount of virus in a medium devoid of nuclei and consequently incapable of developing nuclear inclusions. Other examples of increase in virus in the absence of nuclear inclusions will be given later. Sufficiently delicate cytologic methods have not been applied to ascertain whether the extension by progressive formation of more virus is related to detectable alterations in the medium. Nor have attempts been made to discover whether there are concurrent changes in the functional activity of the nerve fibers by utilization of the cathode oscillographic technic employed by O'Leary, Heinbecker and Bishop<sup>25</sup> for the investigation of physiologic changes in nerve fibers in experimental poliomyelitis. This is a key problem that calls for careful analysis.

22. Goodpasture, E. W., and Teague, O.: J. M. Research **44**:139, 1923.

23. Goodpasture, E. W.: Medicine **8**:223, 1929.

24. de Renyi, G.: Architecture of the Nerve Cell, in Cowdry, E. V.: Special Cytology: The Form and Functions of the Cell in Health and Disease, ed. 2, New York, Paul B. Hoeber, Inc. 1932, vol. 3, p. 1370.

25. O'Leary, J. L.; Heinbecker, P., and Bishop, G. H.: Arch. Neurol. & Psychiat. **28**:272, 1932.

There is evidence that the neurotropic viruses—herpes, poliomyelitis, Borna disease, perhaps others, and rabies, which does not fall in the scope of this paper because it does not produce nuclear inclusions—extend not only to the nervous system in this remarkable manner, but also spread from the nervous system, once they have established themselves there, in the substance of the nerve fibers in a peripheral direction. These particular viruses can be isolated with difficulty or not at all from the blood stream. Other viruses, which do not possess this affinity for nerve tissue, or for which nerve tissue is not selectively vulnerable (which is not exactly the same thing), choose the blood as the means of transport but in varying degrees. Some spread rapidly in it and in large amounts while others remain quite sharply localized in particular tissues or organs. These *differences in mode of spread have not as yet been correlated with the physicochemical properties of the viruses.* Even a beginning has not been made.

*Can the attribute of inclusion formation be correlated with virus properties?*

If size of particle is a measure of diffusibility, it should be feasible, when more data are available, to arrange the viruses in series and perhaps to associate this property with ability to produce inclusions. That the particles are electropositive or electronegative at a certain  $p_H$  also affords a chance of correlation with inclusion formation. Nothing has been done in these directions.

Some viruses are mutants. The change of smallpox virus to vaccinia virus with loss of ability to produce intranuclear inclusions was a mutation. If only more were known of the physical chemistry of these two a clue might be gained to the feature responsible for the formation of inclusions, present in the one and absent in the other. The yellow fever virus in monkeys is hepatotropic, forming inclusions in liver cells. Transfer to the brain of mice renders it neurotropic, with which change it loses the ability to produce inclusions in liver cells and acquires the property of doing so in nerve cells. Again an alteration in virus is definitely related to a change in inclusion production, but it has not been possible as yet to characterize the alteration.

The same virus may yield inclusions in some species and not in others, though it causes disease in both. Two instances may be cited. The virus of louping ill acts in several species in which intranuclear inclusions have not been reported, but they appear in mice.<sup>26</sup> The virus of mad itch produces nuclear inclusions in the rabbit, monkey and cow but not in the domestic pig<sup>27</sup> though the latter may die of the

26. Webster, L. T., and Fite, G. L.: Proc. Soc. Exper. Biol. & Med. **30**:656, 1933.

27. Hurst, E. W.: J. Exper. Med. **58**:415, 1933.

disease. This is important, for it shows that the attribute of inclusion formation can be suppressed by species differences without interfering with the pathogenicity or the increase in the amount of the virus.

The case of virus III is interesting. Rivers and Tillett<sup>28</sup> discovered that by passage of testicle emulsion in series through rabbits this virus made its presence known by an increase in virulence and the development of a marked capacity to produce inclusions. Evidently there was a progressive change in it with the acquisition of this ability, but again from the physicochemical standpoint there is ignorance as to the nature of the alteration.

*Can cell properties be correlated with inclusion formation?*

Species differences in cellular properties commonly control susceptibility to the action of viruses. The control may be second or third hand and dependent on the conditioning by the cells of the tissue fluid or blood plasma. Tremendous doses of a virus easily exciting inclusion formation in one species will fail utterly to produce inclusions or any sign of a specific reaction in a nearly related species. The cell type determines the reaction, but in a less rigid way than the species. Herpes naturally produces inclusions in epidermal cells; mad itch, in nerve cells, and yellow fever, in liver cells. In general, viruses prefer ectodermal and endodermal derivatives and neglect mesodermal ones, but mesodermal cells are not wholly immune. The virus of fox encephalitis is peculiar in its liking for endothelium. Blood cells, muscle, bone and cartilage cells seldom respond to viruses by the development of intranuclear inclusions.

It may or may not be significant that ectoderm and endoderm, either in the embryo or in the adult or in both, line surfaces communicating with the outside. Opportunity for virus action is a factor in the determination of inclusion formation. A potent herpetic virus, such as the H. F. strain, will call forth inclusions when placed in contact with a wide variety of cells in addition to epidermal ones. On the lip it does no particular harm, but in the brain it may kill. The salivary gland virus of guinea-pigs fails to produce a clinically recognizable condition in its home, the salivary glands. When injected into the brain of a young and susceptible guinea-pig it leads to the development of inclusions in nerve cells and macrophages and is often lethal.

No systematic attempt has been reported to relate the alacrity of the formation of inclusions by certain cells and the reluctance or refusal by others to differences in their nuclear structure. At first sight it might appear that those already possessed of more acidophilic nuclear material, like nerve cells, would be more prone to present inclusions.

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28. Rivers, T. M., and Tillett, W. S.: J. Exper. Med. 40:281. 1924.

At the opposite end of the series is the lymphocyte, which has a nucleus the richest of all in basophilic chromatin and which rarely exhibits inclusion formation.

*Can cells be rendered more susceptible to virus action?*

Only isolated experiments have been made with a view to facilitating or inhibiting the response by altering the physiologic state of the cells. Scott<sup>29</sup> found that ligation of gland ducts suppresses, and stimulation with pilocarpine promotes, the formation of nuclear inclusions in the submaxillary glands of guinea-pigs. Having in mind the influence of vitamin B deficiency on nerve cells and the frequency, already cited, with which these cells are affected by viruses, Lucas, Neff and I are trying to measure any difference that may exist in susceptibility to the herpetic virus between rats deficient in vitamin B and normal rats. Thus far the results have been unexpected. The resistance seems to be, not broken down, but slightly enhanced by the deficiency. Chor,<sup>30</sup> working with me, attempted without success to depress natural resistance to the poliomyelitis virus by splenectomy. When investigators do manage to alter the receiving tissue so that this virus "takes," or discover some susceptible animal other than the monkey, one may look for progress comparable with that in yellow fever, which resulted from Max Theiler's transmission of the virus to white mice.

*What cellular changes are coincident with inclusion formation?*

By sifting the inconsequential alterations from the essential ones, one may hope in the end to devise artificial means whereby the development of intranuclear inclusions can be brought about in the absence of virus. Marked hypertrophy of cytoplasm and nucleus is a feature of the influence of some viruses on certain cells. It is most evident in the salivary glands among viruses producing nuclear inclusions. A local osmotic factor is probably involved. When the same viruses act in the brain, hypertrophy is slight. With a number of other viruses it is hardly perceptible. Hyperplasia likewise depends on the particular modes of response of the cells involved. It is sometimes quite extensive in epidermal cells and altogether absent in nerve cells, which have sacrificed the power of multiplication at the altar of extreme specialization. The relation of speed of inclusion formation to the properties of the inclusions has not been analyzed, but is undoubtedly important. As stated, the common denominator in the formation of type *A* inclusions, which are the most typical of virus action, is a change that sweeps through the whole nucleus. It may be electrical, osmotic or both. Perhaps there is a determining or coincident alteration in intranuclear  $p_{H^+}$ .

29. Scott, Gordon H.: J. Exper. Med. **49**:229, 1929.

30. Chor, H.: Arch. Path. **15**:387, 1933.

The injection of indicators might not be feasible because the nuclei are so much smaller than those treated in this way by Chambers. The shifts in material under centrifugal force should be studied in different stages of inclusion formation.

*Is it possible to produce inclusions by means other than viruses?*

The first step in this direction was the observation of Akiyama<sup>31</sup> and of Heinbecker and O'Leary<sup>32</sup> that curious alterations simulating inclusion bodies appear in nerve cells after electrical stimulation. Davenport, Ranson and Terwilliger<sup>33</sup> found that similar changes could be produced by soaking nerve cells in hypertonic salt solutions. They suggested "that the nuclear inclusions observed pathologically may be the result of disturbed osmotic conditions in the cell."

Lee,<sup>34</sup> with the advice of Dr. J. L. O'Leary, carried the experiments much further. He injected strong dextrose solutions intravenously into cats and made preparations of the spinal ganglions at various times thereafter. He encountered the alterations mentioned in the preceding paragraph, unless the animals were allowed to survive more than three hours, in which event no change was seen, proving the temporary nature of this modification in the living animal. It is desirable to emphasize the fact that the change was visible in freshly isolated cells examined in isotonic mediums. In fixed and stained specimens the nuclear inclusions were radically different from those of type *B* and presented certain points of similarity and of dissimilarity to those of type *A*. They resembled the *A* inclusions in that there was a marked increase in acidophilic material, which became heaped up in the center of the nucleus and was separated from the nuclear membrane by a clear halo. They differed from the *A* inclusions (1) by a failure of basophilic chromatin to marginate on the nuclear membrane, (2) by the nucleolus in most instances retaining its central position and (3) by the fact that the process did not go on to complete nuclear degeneration. He then applied a powerful diuretic, salyrgan, both intramuscularly and intravenously, and produced intranuclear inclusions in the pancreas and other glands. Though the nuclei were not so drastically and immediately altered as by viruses, some of the inclusions produced (at from three to thirty days) were indistinguishable from those of type *A*. The nuclei exhibited central acidophilic material, a clear halo and margination of basophilic chromatin with destruction of nucleoli. These experiments are being continued.

31. Akiyama, S.: Arb. a. d. Med. Univ. z. Okayama **1**:278, 1929.

32. Heinbecker, P., and O'Leary, J. L.: Anat. Rec. **45**:219, 1930.

33. Davenport, H. H.; Ranson, S. W., and Terwilliger, E. H.: Anat. Rec. **48**: 251, 1931.

34. Lee, J.: Proc. Soc. Exper. Biol. & Med. **31**:383, 1933.

*Are the viruses which produce nuclear inclusions living organisms?*

The belief that this is the case rests on at least four considerations:

1. Evidence that organisms have been seen in the inclusions. Goodpasture<sup>35</sup> emphasized the occasional basophilia of the inclusions and stated that this is often due to the coloration of small particles of microscopic size, which he has interpreted as elementary bodies—a stage in the life cycle of so-called Chlamydozoa. No proof is, however, forthcoming that the particles are anything other than tiny masses of nucleoprotein which are a little more acid (i. e., more basophilic) than the rest of the inclusion material. Pinkerton and Hass<sup>36</sup> observed that in certain tissue cultures the rickettsiae of Rocky Mountain spotted fever accumulate within nuclei and in their opinion resemble inclusion bodies, but Cowdry pointed out that the correspondence is questionable.

2. These diseases, like those due to bacteria, are transmissible in series, and in each affected individual there is an enormous increase in virus pointing, it is claimed, to multiplication. The strength of this argument in favor of an organismal nature is undeniable unless it should be possible to explain transmissibility and increase in virus on another equally plausible hypothesis.

3. As in an infection due to a living organism, virus diseases do not spring up *de novo* but always appear by extension from preexisting cases.

4. Time and again infectious diseases of long unknown etiology have ultimately been found to be caused by organisms of some sort. But there is no inevitability about it. We are here concerned with those viruses that cause the formation of nuclear inclusions, particularly of type A, not with viruses as a whole. Since none of the viruses thus far proved to be organismal (for example, those of heartwater of sheep<sup>37</sup> and psittacosis<sup>38</sup>) belong to this group the argument loses some weight, but must be borne in mind.

*Are the viruses of this category inanimate substances?*

Chemists are more ready to entertain this concept than bacteriologists. It is a question whether it is more difficult to think of hitherto unrecognized chemical processes or of living things different from those familiar to us.

1. The transmissibility in series and increase in virus during the course of the disease could perhaps be explained on the supposition.

35. Goodpasture, E. W.: Am. J. Path. **1**:1, 1925.

36. Pinkerton, H., and Hass, G. M.: J. Exper. Med. **56**:151, 1932.

37. Cowdry, E. V.: J. Exper. Med. **42**:231, 1925.

38. Lillie, R. D.: Pub. Health Rep. **45**:221, 1930.

purely as a working hypothesis, that the disease inducing viruses of this class are autocatalytic; in other words, that acting on cells they are capable of producing more of the substance of which they are composed.

2. The feature of spread of virus infection from previous cases is not altogether incompatible with the agent being nonliving. It is possible that the virus substance is developed on extremely rare occasions and, once formed, passes from animal to animal, given the opportunity of entry into a mechanically injured cell.

3. But the principal reasons in justification of the hypothesis that these viruses producing nuclear inclusions may be inanimate substances are: (a) They are particulate and much smaller than any known living things. (b) Some of them are not inactivated (or killed) by conditions lethal for living organisms. (c) In the process of chemical purification they do not behave like organisms. (d) The relatively pure product is unstable, like an enzyme. (e) None of them has as yet been cultured on artificial mediums without living cells.

4. To base contentions only on viruses that are transmissible in series and that produce easily recognizable disease is to view only a small corner of the picture. Cytologists are interested in the wide distribution of nuclear inclusions of type *A*. This far exceeds the occurrence of viruses the existence of which has been demonstrated by animal passage. It may be, as already mentioned, that failure to prove virus action means that the required experimental conditions have not been realized. But it is also possible that even in this restricted group of viruses one has to do with substances which are only rarely autocatalytic. On this theory the intranuclear inclusions which are observed so frequently in the total absence of signs of disease and which it is not feasible by present methods to create simply by transfer of tissue emulsion from animal to animal are produced not by a living agent or by an autocatalytic substance but as a result of special local conditions in the respective tissue which one may hope eventually to duplicate experimentally in the absence of a transmissible agent whether organismal or chemical.

#### *What general conclusions can be reached?*

One gains the impression of many data that are poorly correlated. But nuclear inclusions in virus diseases are definite cellular modifications. They can be regarded as the fingerprints of a special and limited group of viruses, which is small in comparison with that hodge-podge of agents to which the term "virus" is ordinarily applied. Moreover viruses, like human beings, may act without leaving fingerprints. The immediacy of relation between virus and inclusions is in question. The

viruses may not themselves produce the inclusions. They may merely initiate physicochemical changes that lead to their production in some cases and not in others. There is a chance that these alterations can be brought about without viruses. What the viruses are, no one knows. There is danger in assuming that they are all alike. Some may be living agents of a type already familiar, or of a type altogether different, while others may be inanimate chemical substances akin perhaps to enzymes.

## Notes and News

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**University News, Promotions, Resignations, Appointments, etc.—** Georges Dreyer, professor of pathology in Oxford University since 1907, died suddenly on Aug. 17, 1934, at the age of 61.

At the University of London, J. R. Marrack has been appointed to the university chair, London Hospital Medical College of Pathological Chemistry.

Carl O. Jensen of Copenhagen, famous on account of his success in transplanting carcinoma in mice in 1901, has died.

W. C. Hueper has been appointed pathologist in the newly founded Institute of Experimental Toxicology of the DuPont Company at Wilmington, Del.

Andrea Saccone and Alfred Aingrist have been promoted to associate professors of pathology in the New York Homeopathic Medical College and Flower Hospital, New York.

Philip B. Hadley, recently associate professor in bacteriology in the University of Michigan, and Arthur P. Locke, recently chief of biochemical research in St. Luke's Hospital, Chicago, have been appointed on the research staff of the Institute of Pathology of the Western Pennsylvania Hospital in Pittsburgh.

Jean Cantacuzène, professor of experimental pathology in the University of Bucharest for more than thirty years, has died. A memorial book in two volumes has been published in his honor by Masson et Cie, Paris.

**Society News.**—The second conference of the International Association for Geographic Pathology was held in Utrecht on July 26 to 28, 1934. The major topic was arteriosclerosis. One session was devoted to hepatic cirrhosis, the subject of the first conference at Geneva in 1931. The third conference will be held in 1937 at a place to be selected. The list of those present at the Utrecht conference contains one hundred and sixty-six names representing twenty countries; ten members of the committee for the United States were present.

**Research in Bacterial Chemistry.**—According to the *Journal of the American Medical Association*, the Medical Research Council in England will sponsor an investigation in bacterial chemistry. Financial provision has been made for an initial period of five years. The work will be carried on under the direction of Paul Fildes at the Middlesex Hospital in the Bland-Sutton Institute of Pathology and the adjoining Courtauld Institute of Biochemistry.

# Abstracts from Current Literature

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## Experimental Pathology and Pathologic Physiology

HYPERACTIVATION OF THE NEUROHYPOPHYSIS AS THE BASIS OF ECLAMPSIA AND OTHER HYPERTENSIVE STATES. H. CUSHING, Am. J. Path. **10**:145, 1934.

From the observations presented the conclusions are drawn: (1) that the source of these hypertensive disorders (eclampsia, essential hypertension) lies in the posterior lobe of the pituitary body; (2) that the extent of basophilic invasion from the pars intermedia is a measure of posterior lobe activity, and (3) that excessive infiltration by these elements represents the histopathologic basis of eclampsia and essential hypertension in young persons and may possibly also be related etiologically to atherosclerosis of old age. Whether the general hypothesis herein advanced should or should not prove on further study to be in all its features wholly correct, it will nevertheless provide an incentive to include a detailed study of the neurohypophysis in forthcoming postmortem studies of disorders in which hypertension is a distinguishing feature.

FROM THE AUTHOR'S CONCLUSIONS.

CARDIOVASCULAR RENAL CHANGES ASSOCIATED WITH BASOPHIL ADENOMA OF THE ANTERIOR LOBE OF THE PITUITARY. H. E. MACMAHON, H. G. CLOSE AND G. HASS, Am. J. Path. **10**:177, 1934.

Two cases of basophil adenoma of the anterior lobe of the pituitary gland, one reported by Bishop and Close and the other by Cushing, have been discussed again from a cardiovascular renal standpoint. It is shown that the cardiovascular renal lesion which was present corresponds to the picture originally described as malignant nephrosclerosis by Fahr.

FROM THE AUTHORS' SUMMARY.

THE CYTOLOGICAL PICTURE OF AN INFLAMMATORY EXUDATE IN RELATION TO ITS HYDROGEN ION CONCENTRATION. V. MENKIN, Am. J. Path. **10**:193, 1934.

The observations reported suggest that the differential leukocyte formula in an area of acute inflammation is a function of the hydrogen ion concentration of the exudate. The cytologic picture seems to be conditioned by the  $p_{\text{H}}$  of the exudate surrounding the injured tissue. The present study indicates that the local acidosis which develops as the inflammatory reaction progresses can adequately account for the shift in infiltration from polymorphonuclear leukocytes to mononuclear phagocytes at the site of inflammation.

FROM THE AUTHOR'S SUMMARY AND CONCLUSIONS.

THE EFFECT OF PARATHYROID HORMONE ON THE CALCIFICATION OF THE DENTIN OF THE RAT INCISOR. I. SCHOUR, W. R. TWEEDY AND F. A. MCJUNKIN, Am. J. Path. **10**:321, 1934.

The principal changes were found in the calcification of the dentin. A primary hypocalcified stripe developed in the dentin that was being calcified immediately after the first injection, and a secondary hypercalcified stripe developed in the dentin that was calcified subsequently. The dentin reaction may depend on control by the parathyroid hormone of a fraction of the serum calcium.

**SELECTION WITH THE MAGNET AND CULTIVATION OF RETICULO-ENDOTHELIAL CELLS (KUPFFER CELLS).** P. ROUS AND J. W. BEARD, *J. Exper. Med.* **59**:577, 1934.

Methods and apparatus are described wherewith living Kupffer cells can be procured from the liver of the rabbit and the dog for study and cultivation in vitro. Almost none of these cells can be dislodged from the normal liver by forcible perfusion; but after they have taken up finely particulate matter (india ink, iron oxide) they come away in great numbers. When they have phagocytosed magnetic iron oxide they can be selected with a magnet from among the blood elements present in suspension with them; and they are obtainable in quantity by this means. They do poorly when plated in a thin plasma clot, failing to multiply or to assume their characteristic shape; but they flourish when allowed to attach themselves to strands of lens paper bathed in serum that is frequently changed. Bacterial infection of serum cultures of Kupffer cells from normal rabbits and dogs occurs only as the result of secondary contamination of the materials, whereas it regularly develops in cultures from animals with fever induced by the injection of nucleic acid or of killed *Bacillus prodigiosus*. Kupffer cells obtained under such conditions are abnormally active, and some can be washed out of the liver of sick animals in the absence of any preliminary phagocytosis of particulate matter. The facts have a bearing both on the conditions conduced to invasion of the blood and on the response of the Kupffer cells in the emergency. The characters of the isolated Kupffer cells and the results of tests of their presumptive functions will be described in later papers.

FROM THE AUTHORS' SUMMARY.

**THE CHARACTERS OF KUPFFER CELLS LIVING IN VITRO.** J. W. BEARD AND P. ROUS, *J. Exper. Med.* **59**:593, 1934.

The Kupffer cells procured from the liver of the rabbit and the dog for culture in vitro have the typical characters of clasmacytes. They are readily discriminated from the monocytes of other organs. Their surface is extraordinarily sticky—far more so than that of blood leukocytes or of the clasmacytes found in peritoneal exudates; and in consequence they are exceedingly difficult to handle in vitro. They put forth enormous, pellucid, circular membranes resembling those of the clasmacytes of exudate but larger. Splenic clasmacytes, on the other hand, put forth rather small, one-sided, ground-glass membranes like broad tongues. On comparing them with Kupffer cells and the clasmacytes of exudate one perceives that they are not wholly identical in their characters, but have secondary peculiarities. However, there exist good morphologic reasons for grouping them together and terming them all reticulo-endothelial. Kupffer cells are notably sensitive to injury, surviving in Tyrode solution for a much shorter time than blood leukocytes. However, they can be readily cultured on lens paper in serum. Under such circumstances they scatter on the fibers and live separately, presenting the same general aspect as when in the liver; but in the course of proliferation they soon lose some of their pronounced characters, retaining such as are common to clasmacytes in general. A considerable population of ordinary leukocytes exists in the hepatic sinuses over and above those circulating in the blood. During infection, their number may greatly increase. Several cubic centimeters of packed white cells have been obtained from the liver of a sick dog. The fact has been realized that leukocytes may stop a while in the liver, yet the extent of the accumulation which sometimes takes place seems deserving of stress.

FROM THE AUTHORS' SUMMARY.

**THE EFFECT OF HISTAMINE ON THE HEALING OF EXPERIMENTAL GASTRIC DEFECTS.** C. A. FLOOD AND E. L. HOWES, *Surg., Gynec. & Obst.* **58**:136, 1934.

The subcutaneous administration of histamine interfered with the healing of mucosal defects in the prepyloric portion of the stomach of the cat and the dog. It delayed but did not prevent healing. Histamine had little effect on the healing of a mucosal defect high on the greater curvature of the stomach. The amount of

histamine required to delay the healing of a prepyloric mucosal defect in the cat for two weeks was from 1 to 1.2 mg. per kilogram twice a day.

#### SUMMARY (W. C. HUNTER).

THE PATHOGENESIS OF HYDRONEPHROSIS. FRANK HINMAN, Surg., Gynec. & Obst. 58:356, 1934.

The pathologic changes of hydronephrotic atrophy are types of the degeneration produced by pressure and anemia and are peculiar to the kidney. Complete obstruction of the excretory ducts of other glands produces almost immediate cessation of function and primary necrosis and atrophy. Primary renal atrophy following complete obstruction of a ureter is the exception and occurs only when there is an initial anuria.

Experimental modification of the secretory pressure does not influence the progressive changes in a related manner. Hydronephrotic atrophy shows the same ordinary rate of development both with water starvation and with forced diuresis. Lowering and raising the pressure by partial obstruction of the renal artery and vein hasten the rate of development, not through the influence of increased intra-pelvic pressure, but because of the increased ischemia.

The degree of the structural repair and restoration of function which follow removal of the obstruction of the ureter varies not only with the extent of the injury which has been produced, which is proportional to the period of obstruction and to the freedom from infection, but also with the degree and manner of the functional stimulation. A gradually increasing excretory load, such as occurs with slow destruction of the opposite kidney, affects a more permanent and greater structural repair than a lesser stimulation, such as occurs when the opposite kidney has undergone compensatory hypertrophy, or a too sudden overload, as in the case of removal of the opposite kidney. An efficient compensatory mate diminishes and an insufficient kidney on the opposite side increases the potentiality of repair of hydronephrosis.

The pathologic changes of hydronephrotic atrophy have been found only in kidneys with internal glomeruli. The gross and microscopic changes occur most typically in the mammalian kidney, which has a hilus and a pelvis. However, this structural arrangement is not essential. Similar changes follow complete obstruction of the ureter or the primary excretory duct of the apelvic kidneys of reptiles, birds and amphibians. The microscopic changes which follow direct obstruction of the tubules in the pelvic type of kidney, such as occurs when a papilla has been tied, resemble those which follow indirect obstruction, such as occurs when the ureter has been tied.

The mechanical factor in the development of hydronephrotic atrophy is a backflow of urine into the venous system. In mammalian kidneys this backflow probably is at first pyelovenous and later tubulovenous. In the apelvic mesometanephroi of reptiles, birds and amphibians the backflow is tubulovenous. Whenever a backflow fails to occur on complete obstruction of the excretory duct, anuria develops, and the pathologic change of primary atrophy instead of hydronephrotic atrophy results.

#### AUTHOR'S SUMMARY (W. C. HUNTER).

DEMONSTRATION OF TRAUMATIC FAT EMBOLISM IN BLOOD AND FATAL AMOUNTS OF FAT. O. SUSANI, Arch. f. klin. Chir. 179:463, 1934.

Susani states that fat embolism is caused by neutral fat, principally olein. For this reason, determinations of the total lipoids or of partial lipoids are of no diagnostic value. The normal neutral fat values as given in the literature are frequently grossly erroneous. He presents exact methods for determining the presence and the amount of neutral fat in the peripheral blood. In severe fractures considerable amounts of neutral fat may be demonstrated in the peripheral blood, frequently associated with shock. This increase is considered a manifestation of latent fat embolism. Alimentary hyperlipemia is a definite entity in which the

values of blood fat may far exceed the normal. The fatal amount of fat is dependent on the state of division of the fat. In a moderately coarse state of comminution smaller fractions of fat may be more fatal than they would be if they were in the crude state. Pure fat induces death from a cardiac failure, whereas emulsified fat is fatal because of asphyxiation.

EXPERIMENTAL GLOMERULONEPHRITIS. W. MASUGI, *Beitr. z. path. Anat. u. z. allg. Path.* **92**:429, 1934.

This work on rabbits was a continuation of previous work on glomerulonephritis in rats from the intravenous injection of antikidney serum. In the rabbit the clinical manifestations, the urinary findings, the retention of nitrogen in the blood and the structural changes of the kidney were identical with those considered characteristic of chronic diffuse glomerulonephritis in man. The reaction in the kidney is held to be an allergic one.

O. T. SCHULTZ.

EXPERIMENTAL HYPERERGIC CARDITIS AND ARTERITIS. E. JUNGHANS, *Beitr. z. path. Anat. u. z. allg. Path.* **92**:467, 1934.

Swine serum, which was used in Junghans' experiments, is toxic to the rabbit and evokes a local reaction at the point of first injection. Ten rabbits were sensitized to swine serum by from three to six subcutaneous injections of from 0.5 to 2 cc. each at intervals of from six to nineteen days. After sensitization the animals were given one or two intravenous injections of swine serum in amounts varying from 9.25 to 10 cc. Five of the animals received from three to five intravenous injections at varying intervals, with a maximum single dose of 20 cc. The animals were killed and the heart, aorta and pulmonary artery examined microscopically. The heart valves revealed fibrinoid swelling of the connective tissue and cellular proliferation; the walls of the coronary arteries were swollen and surrounded by granulomatous perivascular inflammation, and the muscle fibers were damaged. This series of reactions Junghans terms "hyperergic carditis." In the aorta and pulmonary artery the media was swollen, and all the coats contained areas of cellular infiltration. The vascular reaction is termed "hyperergic arteritis." It is identical with the arterial changes noted in rheumatic infection.

O. T. SCHULTZ.

WEATHER CHANGES AND APPROACHING DEATH. G. ORTMANN, *Virchows Arch. f. path. Anat.* **291**:237, 1933.

This is an investigation similar to that made by Struppner in Munich, which has been previously abstracted (*ARCH. PATH.* **15**:280, 1933). It is based on more than 16,000 deaths that came to necropsy in Berlin. As in Munich, there were noted periodic increases in the number of deaths. These fluctuations were correlated with changes in meteorological conditions. Such a study appears to have value only if the deaths submitted to necropsy in a large hospital represent all or an overwhelming proportion of the deaths that occur in the institution. Ortmann found that there was a distinct increase in deaths on those days when there occurred a change in weather due to the passage of a cold front or a warm front over Berlin.

O. T. SCHULTZ.

DEVELOPMENT OF COLLAGEN FIBRILS IN TISSUE CULTURE. L. DOLJANSKI AND F. ROULET, *Virchows Arch. f. path. Anat.* **291**:260, 1933.

The authors trace the development of fibrils with the staining reactions of collagen in the plasma medium of tissue cultures. A transformation of fibrin into collagen was observed, but the formation of collagen fibrils may and does occur independently of the fibrin network of the plasma coagulum. The fibrils stain by the Foot silver method. They are most numerous immediately about the cell

bodies, but were never observed within the cell body or in the elongated processes of the mesenchymal cell. Although they arise independently of the cell, their formation occurs under the influence of cellular activity. The cell exerts its influence on fibril formation in the plasma medium by means of a secreted material, possibly something in the nature of an enzyme. Low diffusibility of the secreted material is inferred from the greater degree of fibril formation immediately about the cells of the culture.

O. T. SCHULTZ.

**RELATIONSHIPS OF THE THYMUS TO THE ENDOCRINE AND LYMPHATIC SYSTEMS.**  
HANNA SCHULZE, *Virchows Arch. f. path. Anat.* **291**:461, 1933.

Injection of thyroxine into young animals, especially mice, led to atrophy of the thymus. The atrophy was due to a direct local destruction of lymphocytes and to increased emigration of lymphocytes from the thymus. In young guinea-pigs thymectomy regularly resulted in compensatory hypertrophy of the cervical lymph nodes and occasionally in hypertrophy of the spleen. The author interprets these findings as proof of a relationship of the thymus to both the endocrine and the lymphatic system.

O. T. SCHULTZ.

**THE EFFECT OF HIGH TEMPERATURE ON THE ALIMENTARY TRACT AND KIDNEY.**  
J. F. BRODSKY, *Virchows Arch. f. path. Anat.* **291**:589, 1933.

In this experimental study of occupational disease, sixteen dogs and three cats were subjected for variable periods to high environmental temperatures such as prevail in certain occupations. Diarrhea developed, as it does in workers in heated atmospheres. In the large intestine there were noted degeneration and desquamation of the superficial epithelium. Similar degenerative and desquamative changes were noted also in the pancreas and the kidney. In the more prolonged experiments fibrosis of the pancreas developed. Spivack had ascribed the diarrhea of workers in heated environments to excessive water drinking forced by thirst caused by loss of water from the body. The experimental animals did not consume unusual quantities of water. Brodsky ascribes the clinical manifestations and pathologic changes noted by him to intoxication. He recommends that workers in heated environments be subjected to examination every three to six months, especial attention being paid to the urine to detect renal damage.

O. T. SCHULTZ.

**THE INFLUENCE OF PHOTODYNAMIC SUBSTANCES ON THE BLOOD LEUKOCYTIC PICTURE OF RABBITS.** W. N. NEKLUDOW, *Virchows Arch. f. path. Anat.* **291**:600, 1933.

Quinine hydrochloride or eosin in solutions of 0.1 and 0.01 per cent respectively was injected intravenously into rabbits and a study of the leukocytes of the peripheral blood made at varying intervals. In some experiments the solutions were subjected to ultraviolet irradiation before injection; in others the animals themselves were irradiated just before or just after the substance was injected. The solutions used caused temporary leukopenia followed by leukocytosis, with a shift to the left and an increase in pseudo-eosinophils. These changes were more marked if the solutions or the animals were irradiated.

O. T. SCHULTZ.

**EXPERIMENTAL FETAL INFLAMMATION.** F. WOHLWILL AND H. E. BOCK, *Virchows Arch. f. path. Anat.* **291**:864, 1933.

Previous study of human placentitis and fetal infection had established that the reaction of the fetal tissues is chiefly histiocytic. In this experimental study of fetal inflammation turpentine or suspensions of bacteria or of india ink were injected into the amniotic liquor of guinea-pigs or into the umbilical cord, skin,

muscles or internal organs of the fetuses. The injections were made at various stages of pregnancy. Fetal sepsis was most often bronchiogenic, as had been previously observed in the human material, and resulted from the aspiration of infected amniotic fluid. A morphologic reaction to bacterial invasion was not observed in the fetuses until near the end of gestation and was macrophagocytic. A histiocytic reaction to turpentine and india ink was observed early in pregnancy and was similar to that which occurs in coelenterates. Even after development of the circulatory system the reaction was primarily histiocytic. Late in gestation a few cells of myeloid origin appeared, but this phenomenon was always preceded by a histiocytic reaction.

O. T. SCHULTZ.

STUDIES ON [HUMAN] GONADOTROPIC HORMONES FROM THE HYPOPHYSIS AND CHORIONIC TISSUE WITH SPECIAL REFERENCE TO THEIR DIFFERENCES. C. HAMBURGER, *Acta path. et microbiol. Scandinav.*, supp. 17, 1933, p. 1.

The studies reported have been limited to the effect on the ovary of gonadotrophic hormones of human origin. Infantile mice and rats have been used for the tests. A main result of the work is that the stimulation of the ovary by hypophyseal hormone (urine of castrates) differs in type from that by hormones from chorionic tissue (urine of pregnant women).

### Pathologic Anatomy

RUPTURE OF THE RIGHT AURICLE OF THE HEART. G. M. CLOWE, E. KELLERT AND L. W. GORHAM, *Am. Heart J.* 9:324, 1934.

A case of apparently spontaneous rupture of the right auricle is reported in a supposedly normal person in whom trauma and excessive exertion were absent. Serial electrocardiograms showing changes suggestive of coronary occlusion are presented for the first time. Histologic study revealed a preexisting obliterating endarteritis with interstitial hemorrhage and infarction of the auricular wall, leading to rupture, a process essentially similar to that causing rupture of the ventricle. The clinical and pathologic features of rupture of the auricle based on a review of fifty-four cases in the literature, plus the one described here, are discussed. The large incidence of rupture of the auricle before the fortieth year of life, 47.7 per cent, as against 6.7 per cent for ventricular rupture, is pointed out.

#### AUTHORS' SUMMARY.

PARADOXICAL EMBOLISM OF THE CORONARY ARTERY. J. JACOBI AND OTHERS, *Am. Heart J.* 9:414, 1934.

A case of acute coronary occlusion due to an embolus from an old thrombosis of the femoral vein in the presence of a patent foramen ovale is reported. There were no associated arterial or endocardial changes. The factors in its production are discussed briefly.

#### FROM THE AUTHORS' SUMMARY.

CORONARY ARTERIES IN RHEUMATIC FEVER. H. T. KARSNER AND F. BAYLESS, *Am. Heart J.* 9:557, 1934.

Rheumatic fever regularly produces disease of the coronary arteries. Either inflammatory or fibrotic lesions or both are practically constant. Except for participation by Aschoff nodules, the lesions are not specific for rheumatic fever. Fibrinoid degeneration is suggestive but not diagnostic. Elastica degeneration appears to be especially severe. The coronary disease is irregularly distributed as to both the various divisions of the coronary tree and the individual members affected. Its relation to myocardial disease cannot be positively established, but the late myocardial fibrosis is greater than is to be expected from the early acute

myocarditis alone. The influence of the coronary disease on myocardial fibrosis is better explained by intimal fibrosis than otherwise. Rheumatic fever predisposes to fibrosis of the coronary arterial tree in early life and to what appears to be precocious coronary sclerosis; but, although this is probably a chronic inflammation, it has not been shown conclusively to be dependent on the acute degenerative and inflammatory lesions. The coronary arteries in rheumatic fever undergo a progressive sequence of inflammatory lesions which closely resemble those of the endocardium and pericardium. It is practically certain that severe myocardial damage is associated with the arterial disease. The resulting effect on myocardial efficiency appears to be of significance in the clinical management and prognosis of rheumatic heart disease.

FROM THE AUTHORS' CONCLUSIONS.

HODGKIN'S DISEASE OF THE LUNG. S. E. MOOLTON, Am. J. Cancer **21**:253, 1934.

Study of the lesions in the lungs in eight cases of Hodgkin's disease leads Moolten to emphasize that such study concerns a primary inflammatory process of granulomatous nature rather than a neoplasm. The uniform infiltration of the structures suggests a diffusible virus or toxic substance. A "primary pleurogenous" form of Hodgkin's disease of the lung is described, apparently for the first time. In most cases a peribronchial interstitial pneumonia is the main lesion, associated with more or less extensive granulomatous parenchymatous pneumonia. In about 10 per cent of all cases of Hodgkin's disease in the lungs the primary lesion is in the lung.

THE RENAL LESIONS OF RHEUMATIC FEVER. J. L. BLAISDELL, Am. J. Path. **10**:287, 1934.

In a study of the kidney in sixteen cases of rheumatic fever, a perivascular inflammatory reaction of the acute nonsuppurative type, affecting the smaller arteries and arterioles, was present in eight cases. Evidence of perivascular scarring was noted in four cases, while a recurrent inflammation was met with in two. The inflammatory reaction is usually seen in the adventitia and peri-adventitial tissues, with occasional infiltration and destructive change in the medial coat. Intimal changes, consisting of an endothelial swelling and proliferation, are constant. Glomerular damage, which was well marked in only one case, is to be regarded as dependent chiefly on nutritional disturbances brought about by the vascular changes. Little evidence of active or healed inflammatory processes was met with in the glomeruli. No evidence of the specific vascular lesions described by Pappenheimer and Von Glahn was encountered in the cases studied. The lesions described, which in general bear a close resemblance to perivascular foci of inflammation found in the myocardium, may be looked on as constituting a definite type of interstitial nephritis. It is seldom, however, that sufficient alteration in structure occurs to justify a diagnosis of renal disease during life.

FROM THE AUTHOR'S SUMMARY AND CONCLUSIONS.

MICROGLIA-LIKE CELLS IN THE LIVER, SPLEEN AND KIDNEY. H. S. DUNNING AND L. STEVENSON, Am. J. Path. **10**:343, 1934.

By del Rio Hortega's original silver carbonate method of specific staining for microglia cells have been demonstrated in the liver, spleen and kidney of the rabbit which in morphology are identical with the nearly normal or very early transitional forms of microglia in the nervous system. In their reaction to injury and to intravital injections of trypan blue they have been shown to be identical with microglia. These cells have been demonstrated in a transitional stage with spiked processes like microglia and containing droplets of fat or granules of trypan blue. By the silver carbonate method of staining earlier transitional forms have been demonstrated that contain no visible amounts of fat or trypan blue. A more advanced

transitional form has been shown in preparations of the spleen of the rabbit to be a histiocyte or large mononuclear phagocyte without processes and containing droplets of fat, granules of trypan blue, blood pigment and engulfed lymphocytes.

## FROM THE AUTHORS' SUMMARY.

**POLYARTERITIS NODOSA.** R. B. HAINING AND T. S. KIMBALL, Am. J. Path. **10**:349, 1934.

The inflammatory changes are not confined to the adventitia and periarterial tissue as indicated by the name "periarteritis nodosa." On the contrary all the vascular coats may be involved. The primary changes may take place in the media, resulting in aneurysm. Involvement of the intima may result in thrombosis. The name "polyarteritis nodosa" is suggested as a better term.

**HISTOLOGICAL CHANGES IN THE CENTRAL NERVOUS SYSTEM FOLLOWING EQUINE ENCEPHALOMYELITIS.** O. LARSELL, C. M. HARING AND K. F. MEYER, Am. J. Path. **10**:361, 1934.

The most constant feature is perivascular infiltration. There may be degeneration of the Nissl substance and necrosis of nerve cells. Cytoplasmic inclusions are found in nerve cells.

**TRACHEO-ESOPHAGEAL FISTULA OF SYPHILITIC ORIGIN.** C. J. BUCHER AND J. ONO, Am. J. Path. **10**:391, 1934.

A case of tracheo-esophageal fistula of syphilitic origin in a 42 year old Negress is reported. Other lesions were gummas of the liver and lymph nodes and incipient aneurysm of the aorta.

## FROM THE AUTHORS' SUMMARY.

**AMYLOIDOSIS IN TUBERCULOUS RABBITS.** R. M. THOMAS, Am. J. Path. **10**:419, 1934.

Amyloid degeneration occurred in 52 per cent of 175 rabbits experimentally infected with bovine tubercle bacilli. The occurrence of amyloidosis was restricted to animals surviving longer than two months after infection. The frequency of occurrence was greatest after the eighth month (75 per cent). The organs affected were the spleen, liver and kidneys, the spleen being most frequently affected. There was a uniform tendency for the deposition of amyloid to occur in those animals that showed the most extensive caseation of their lesions.

## FROM THE AUTHOR'S SUMMARY.

**THE ADRENAL CORTEX IN MONGOLISM.** L. C. HIRNING AND S. FARBER, Am. J. Path. **10**:435, 1934.

In mongolian idiots, as maturity advances, a definite hypoplasia of the suprarenal cortex becomes evident by the use of histologic methods in the measurement of the width of the permanent cortex of the suprarenal gland.

## FROM THE AUTHORS' CONCLUSIONS.

**SARCOID OF BOECK (BENIGN MILIARY LUPOID) AND TUBERCULIN ANERGY.** MARION B. SULZBERGER, Am. Rev. Tuberc. **28**:734, 1933.

Sulzberger reviews the classification of cutaneous tuberculous manifestations into three more or less well defined groups, tuberculosis, the tuberculids and the sarcoid group suggested by Lewandowsky, Jadassohn and Martenstein. In a woman with tuberculous sarcoid of Boeck and typical sarcoid infiltration of the lungs there was a lasting hyposensitivity or anergy to tuberculin. Microscopically there were naked epithelioid cell tubercles without many lymphocytes and without

necrosis or caseation. No bacilli could be demonstrated. It is interesting to note that Boeck's sarcoid not infrequently affects other organs. Next to the pulmonary changes, the most frequent are the cystic changes in the phalangeal bones (absent in this case). Sometimes there are sarcoids of the spleen and liver and even a sarcoid-iritis.

H. J. CORPER.

GENERALIZED THROMBO-ANGIITIS OBLITERANS. W. BIRNBAUM, M. PRINZMETAL AND C. L. CONNOR, Arch. Int. Med. **53**:410, 1934.

An unusual case of generalized vascular disease is reported, probably a case of early thrombo-angiitis obliterans, in which autopsy was possible early in the course because of the involvement of vital structures (infarction in the suprarenal glands). Involvement of the cerebral, retinal, pulmonary, coronary, mesenteric, suprarenal, pancreatic, duodenal, hepatic, renal and prostatic vessels and of the vessels of the extremities was found. The possibility that thrombo-angiitis obliterans is more frequently a generalized disease is pointed out.

#### AUTHORS' SUMMARY.

STRUCTURAL CHANGES IN THE BRAIN FROM TRAUMA. N. W. WINKELMAN AND JOHN L. ECKEL, Arch. Neurol. & Psychiat. **31**:956, 1934.

Injuries to the brain cause a number of gross and microscopic cerebral lesions depending on the extent and the degree of the trauma. The authors attempt to describe the central nerve changes in seven cases which they also studied clinically. The most outstanding changes were subarachnoid bleeding with reactive pial phenomena which may ultimately result in pial-cortical adhesions; intracerebral hemorrhages with "maceration" of the brain tissue; occasional edema which, when present, causes general ischemia with ganglion cell changes, and generalized gliosis which tends to "disappear gradually." The foregoing changes among others less common suffice to explain the neurologic and mental pictures in severe cases. According to the authors, changes in the brain are also the underlying cause of so-called traumatic neuroses.

GEORGE B. HASSIN.

SPLENITIS DUE TO UREMIA. K. VON WOLFF, Beitr. z. path. Anat. u. z. allg. Path. **92**:230, 1933.

Von Wolff describes changes in the spleen in uremia which he considers inflammatory. He divides his material into four groups: (1) cases with an increased number of leukocytes and proliferated pulp cells in the spleen; (2) those with serous, hemorrhagic and occasionally fibrinous exudate; (3) those with reticulo-endothelial proliferation, and (4) those with necrosis and necrobiosis. These various reactions are ascribed to the toxic action of protein degradation products on the spleen.

O. T. SCHULTZ.

CHANGES IN THE EXTRINSIC EYE MUSCLES IN THYROTOXICOSIS. E. VON ZALKA, Beitr. z. path. Anat. u. z. allg. Path. **92**:239, 1933.

The external eye muscles of sixteen persons who died of exophthalmic goiter were examined histologically. Severe changes were noted. These included proliferation of sarcoplasm (Durante's cellular regression), lymphocytic infiltration and lipomatosis. Proliferation of sarcoplasm was seen in other conditions, but was less marked than in thyrotoxicosis. Lymphocytic infiltration is secondary to degenerative changes in the muscle and is not part of the lymphatism so frequently seen in exophthalmic goiter. The alterations described are held to be the result of the increased functional activity of the disease. They develop as the result of exophthalmos but may be a factor in bringing about an increasing degree of this condition.

O. T. SCHULTZ.

PITUITARY BASOPHILISM. E. RUTISHAUSER, Deutsches Arch. f. klin. Med. 175:640, 1933.

In three cases of osteoporotic obesity there was an increase in the basophil cells of the anterior lobe of the pituitary gland.

CONGENITAL SYPHILITIC KIDNEY WITH CARDIAC HYPERSTROPHY AND CEREBRAL HEMORRHAGE IN AN EIGHT YEAR OLD CHILD. H. LUDTKE, Frankfurt. Ztschr. f. Path. 44:405, 1933.

A child, aged 8, died from cerebral hemorrhage. The genesis of the contracted kidneys is traced to a syphilitic renal endarteritis. While the cardiac hypertrophy must be attributed to the contracted kidneys, the cerebral hemorrhage is considered due to both the high blood pressure and the syphilitic changes of cerebral blood vessels.

WILLIAM SAPHIR.

FATAL PULMONIC CIRRHOsis PRODUCED BY MENTHOL. B. FISCHER-WASELS, Frankfurt. Ztschr. f. Path. 44:412, 1933.

The autopsy on an 86 year old woman who gave the clinical picture of a progressively malignant pulmonic tumor revealed a massive tumor-like fibrosis involving regions about the hilus in both lungs. Histologic examination showed a dense scarlike connective tissue formation with numerous fat droplets. By histochemical examination these fat droplets were determined to be paraffin oil. Subsequently it was learned that the patient had suffered from a chronic nasopharyngitis and had used for more than twenty years, a 1:1,000 solution of menthol as nose drops. It must be assumed that the anesthetizing property of menthol and the chronic bronchitis favored the entrance of the oil into the bronchial tubes and alveoli. The presence of the oil in the tissue, acting on them as a foreign body, finally gave rise to the formation of the marked fibrosis.

WILLIAM SAPHIR.

CONGENITAL EPIDERMOID CYST OF THE HEART. A. DE CHATEL, Frankfurt. Ztschr. f. Path. 44:426, 1933.

In a new-born girl, a cyst of the interauricular septum was found, which on histologic examination appeared to be a multilocular dermoid cyst containing derivatives of the three germinative layers. Chatel was unable to find record of a similar case in the literature. He is inclined to explain the dermoid cyst on the basis of misplaced ectodermal cells or on that of a metaplasia of entodermal cells in a very early stage of development. A meningo-encephalocele and a defect of the nasal septum were also found at autopsy.

WILLIAM SAPHIR.

TWO RARE HEMORRHAGES INTO THE ESOPHAGUS AND STOMACH. S. SCHEIDEGER, Frankfurt. Ztschr. f. Path. 44:527, 1933.

In one instance the hemorrhage was due to ruptured esophageal varicose veins the condition of which was attributed to an old thrombosis of the portal vein. It seems most probable that the thrombosis was the result of a severe appendicitis which occurred in childhood. In the other instance the fatal hematemesis was due to a congenital anomaly of submucous gastric arteries, which were very wide and tortuous. Following a meal one superficial branch became damaged and ruptured, with the subsequent fatal hemorrhage. In both patients the changes apparently had been present for many years but had not given rise to clinical symptoms.

WILLIAM SAPHIR.

BLIND ARTERIES WITH INVAGINATION OF THE INNER COATS IN THE REGION OF THE DUCTUS ARTERIOSUS. E. FRITZ, Frankfurt. Ztschr. f. Path. 45:273, 1933.

In a new-born infant who had lived four and one-half hours a small blind artery was found in the loose connective tissue between the ductus arteriosus and the left bronchus. Close to the distal end of the artery its lumen revealed inverted portions of intima and media, which in some sections appeared free within the lumen, seemingly not attached to the wall. Another artery, also with inversion of its intima and parts of the media, was found between the ductus arteriosus and the left bronchus. The walls of the vessel revealed no pathologic changes. The belief is expressed that the inversion of the inner coats might have been the result of trauma which, however, could not be demonstrated. The absence of hemorrhage or any inflammatory changes speaks against birth trauma as the etiologic factor.

O. SAPHIR.

ECHINOCOCCUS DISEASE IN THE HEPATIC DUCTS. W. S. SKLJANIK, Frankfurt. Ztschr. f. Path. 45:278, 1933.

Two instances are described. In both the exact diagnosis was made at autopsy. Clinically, cholecystitis and choledolithiasis with obstruction of the hepatic ducts were diagnosed in one instance, and possible pancreatic tumor with subdiaphragmatic abscess, in the other. Both patients were markedly jaundiced. In both instances the involvement of the hepatic ducts by the echinococci was secondary, the liver having been involved primarily. The author concludes that in instances of sudden obstruction of the bile ducts combined with diarrhea the feces should be examined for echinococci. He also believes that an intermittent appearance of a tumor in the region of the liver is pathognomonic of echinococci involving the hepatic ducts. In every disease of the hepatic ducts combined with sudden obstruction of the ducts, echinococcus disease should be considered.

O. SAPHIR.

VARICOSE VEINS IN THE SMALL AND LARGE INTESTINES CAUSING FATAL HEMORRHAGE. F. FENSTER, Frankfurt. Ztschr. f. Path. 45:316, 1933.

At the autopsy on a 56 year old man who had bloody stools a varicosity was found in the submucous veins of the ileum and colon. One of the veins had ruptured and caused the fatal hemorrhage. The veins showed a partial loss of the media. The elastic fibers, particularly, were absent. A primary malformation of the elastic and muscular constituents of the walls of the veins seemed the best explanation for the formation of the varicosity.

O. SAPHIR.

TOXIC LESION OF THE BRAIN IN EARLY INFANCY. F. HOLLINGER, Frankfurt. Ztschr. f. Path. 45:346, 1933.

The literature of so-called fetal encephalitis is discussed. This disease, with the exception of a syphilitic meningo-encephalitis, is rare. An intra-uterine infection plays apparently no rôle in its causation. Five cases are described. In three the Wassermann test of the infants and mothers, respectively, was positive. The encephalitis was attributed to the therapeutic use of sulpharsphenamine. In one infant microgyria and porencephalia were thought to be the result of the encephalitis. In the other two infants the encephalitis seemed to be the result of a gastrointestinal intoxication.

O. SAPHIR.

RHEUMATIC PERIVASCULAR SCARS OF THE MYOCARDIUM. F. WILD, Virchows Arch. f. path. Anat. 290:116, 1933.

The fourteenth in the series of contributions by Klinge and his associates on the pathology of rheumatic infection is based on microscopic examination of the

139 hearts studied in the preceding communication (abstr., Arch. Path. 17:113, 1934), to determine the relation of arteriosclerosis to rheumatism. The same classification of the cases, based on clinical and gross anatomic data, is used in this article. The lesion with which Wild deals is the end-stage of the rheumatic granuloma or Aschoff body; he considers it histologically characteristic of antecedent rheumatic infection. It is an oval or spindle-shaped lesion, sometimes just barely visible to the naked eye, situated about the smaller arteries of the myocardium. It consists of dense, often hyaline, fibrous tissue, which may be arranged about the vessel like the layers of an onion. It is the result of characteristic damage to connective tissue during the acute stage of the infection and is not a replacement fibrosis of primarily damaged muscular fibers. This lesion, Wild believes, may readily be distinguished from senile myocardial fibrosis, which is also perivascular, and from the interstitial fibrosis of coronary sclerosis, syphilis and other diseases. Wild's findings were as follows, the first figure being the number of cases in the group and the figure in parentheses the percentage with rheumatic scars: active rheumatic infection at the time of death, 5 (100 per cent); known previous rheumatic infection with endocarditis, 8 (87.5 per cent); probable rheumatic infection with endocarditis, 7 (71.4 per cent); endocarditis without clinical history or gross evidence of rheumatism, 20 (45 per cent); syphilis, 15 (40 per cent); chronic infection, 10 (40 per cent); arterial hypertension, 22 (22.7 per cent); acute infection, 13 (53.8 per cent); sepsis, 3 (33.3 per cent); chronic tuberculosis, 10 (40 per cent); miscellaneous, 26 (46.1 per cent). Such high percentages in diseases other than rheumatism might lead to the thought that the lesion which is held by Wild to be pathognomonic of rheumatic infection is not specific. Wild maintains that his figures are minimal ones and that they indicate the great frequency of rheumatic infection in and about Leipzig.

O. T. SCHULTZ.

**ENCEPHALITIS ASSOCIATED WITH ARTERITIS.** FRANCIS HARBITZ, Acta path. et microbiol. Scandinav., supp. 16, p. 101, 1933.

Attention is called to some rare forms of arteritis accompanied by confusing nervous reactions, the correct diagnosis being reached only after microscopic examination. A woman of 39 complained of double vision, strabismus, protrusion of the right eyeball and drowsiness. There was found: thrombophlebitis of the cavernous and superficial petrosal sinuses associated with encephalitis of the temporal lobe and arteritis. A 42 year old man who had suffered for ten years from periodic attacks of headache on the right side was found to have arteritis with secondary necrosis and inflammation in the right cerebral hemisphere. No bacteria were demonstrable in the inflamed vessels.

JACOB KLEIN.

**AMYLOID DEGENERATION IN ANIMALS.** A. HJÄRRE, Acta path. et microbiol. Scandinav., supp. 16, p. 132, 1933.

A review of amyloid degeneration in various animals is presented. The liver and spleen are particularly susceptible to amyloidosis in horses, birds and cats; the kidneys, in cattle and dogs. There is a severe form of amyloidosis of the skin and lymph glands in the horse. Generalized amyloidosis occurs in animals especially after chronic infections (in the horse after immunizing procedures in obtaining antitoxic serum).

JACOB KLEIN.

**HEREDITARY DWARFISM IN THE MOUSE.** TAGE KEMP, Acta path. et microbiol. Scandinav., supp. 16, p. 189, 1933.

Hereditary dwarfism in the mouse has been found in a strain brought from England. It behaves in inheritance as a recessive mendelian character depending on a single gene. Some of the endocrine glands in the dwarf are abnormal and

reduced in size—the thyroid gland, the suprarenal cortex, the gonads and the pituitary gland. The chief cause of the condition is considered to be a hereditary deficiency of the anterior lobe of the pituitary gland.

JACOB KLEIN.

**TOTAL GANGRENE OF THE GASTRIC MUCOSA IN GRANULOPENIA.** G. FALK and N. GELLERSTEDT, Acta path. et microbiol. Scandinav., supp. 16, p. 50, 1933.

In a woman, aged 50, with a diagnosis of aleukemic lymphadenosis, roentgen treatments were followed by a marked reduction of the granulocytes in the blood and death. The entire lining of the stomach was gangrenous.

**RETICULO-ENDOTHELIOSIS.** M. G. NORDENSON, Acta path. et microbiol. Scandinav., supp. 16, p. 255, 1933.

In a woman, aged 65, who died with severe anemia after an illness of about two and one-half months, there was found some enlargement of the liver, spleen and some of the lymph nodes; also, marked osteoporosis with red marrow in the femur and vertebrae. Microscopically there was a general proliferation of the reticulo-endothelial system in the liver, spleen, marrow and lymph nodes.

### Pathologic Chemistry and Physics

**BILE CHOLESTEROL.** A. WRIGHT AND G. H. WHIPPLE, J. Exper. Med. 59:411, 1934.

Under uniform conditions of diet the normal dog with a bile fistula will eliminate fairly constant amounts of cholesterol—about 0.5 to 1 mg. per kilogram of body weight every twenty-four hours. Diets rich in cholesterol (egg yolk) raise the output of cholesterol in the bile, but the increase is trivial (from 5 to 15 mg.) compared with the intake (1.5 Gm.). Calves' brains in the diet are inert. Bile salt alone will raise the cholesterol output in the bile as much and often more than a cholesterol-rich diet. Bile salt plus egg yolk plus whole bile gives the maximal output of biliary cholesterol—60 mg. each twenty-four hours. Hepatic injury (chloroform) decreases both the bile salt and the cholesterol in the bile. Blood destruction (hydrazine) fails to increase the cholesterol output in the bile, and this eliminates the red cell stroma as an important contributing factor. Certain cholagogues (isatin and sodium dehydrocholate) increase the bile flow but cause no change in the cholesterol elimination. The ratio of cholesterol to bile salt in the bile normally is about 1:100, but the bile salts are more labile in their fluctuations. The ratio is about reversed in the circulating blood plasma, where the cholesterol is high (from 150 to 300 mg. per hundred cubic centimeters) and the bile salt very low. In the bile cholesterol runs so closely parallel to bile salt that one may feel confident of a physical relationship. In addition there is a suspicion that the cholesterol of the bile is in some obscure fashion linked with the physiologic activity of the hepatic epithelium.

#### FROM THE AUTHORS' SUMMARY.

**BLOOD PLASMA CHOLESTEROL.** W. B. HAWKINS AND A. WRIGHT, J. Exper. Med. 59:427, 1934.

Hypocholesterolemia with dissociation of the normal ratio of esterified to total cholesterol is related to chronic hepatic injury caused by chloroform. Hypercholesterolemia may develop after prolonged biliary obstruction. Such hypercholesterolemia may be promptly reduced below normal by chloroform poisoning or by an infection of the bile duct. Acute injury of the liver following chloroform anesthesia may cause no change in blood plasma cholesterol. Absence of bile in the intestine with faulty absorption of fat does not cause hypocholesterolemia with dissociation of the ester ratio. Inadequate consumption of food or short periods of

fasting may cause no change in blood plasma cholesterol. Liver cells injured by chloroform may subsequently become resistant to chloroform. After prolonged biliary obstruction the liver is apparently more sensitive to small doses of chloroform by mouth. Analysis of blood plasma cholesterol may have a clinical application in differentiation between simple obstructive and parenchymatous lesions of the liver.

## FROM THE AUTHORS' SUMMARY.

THE EFFECT OF CARROT FEEDING ON THE SERUM PROTEIN CONCENTRATION OF THE RAT. A. L. BLOOMFIELD, *J. Exper. Med.* **59**:687, 1934.

It has been shown that rats subsisting on a diet of carrots remain in good condition for periods of as long as twenty-one weeks. There are, however, loss of weight, at first rapid, later more gradual, and a fall in the serum proteins. When this drop is extreme (4.5 Gm. or more per hundred cubic centimeters) ascites and hydrothorax are likely to develop. The response of the individual rats varies greatly, however, even when they are all maintained under similar conditions, so that loss of weight and drop in serum proteins occur much more rapidly in some animals than in others. In the interpretation of the experiments, the point at issue is whether carrots contain an agent which has a disturbing effect on the mechanism regulating the concentration of the serum proteins or whether the drop in serum proteins is a nonspecific effect of malnutrition. The fact that controls on a variety of low protein, ill-balanced, vitamin-deficient diets failed to have edema and suffered for the most part only a slight, if any, drop in serum proteins below the physiologic level suggests that carrots exercise a deleterious influence. Further analysis has shown, however, that a diet of dried carrot powder leads neither to hypoproteinemia nor to edema (ascites), whereas the forced addition of water by suspending the powder in agar reproduces all the effects of fresh carrots. Water, therefore, seems to be the crucial factor rather than some specific constituent of carrots. Incidentally it may be noted that the artificial carrot offers a simple and certain method of producing hypoproteinemia in an animal otherwise in good nutritive condition, and it is proposed to use this technic for the study of further aspects of the subject. Finally it is of interest to correlate the carrot hypoproteinemia with clinical malnutritional disorders. It appears that a combination of factors is necessary in both cases, namely, an intake inadequate in total number of calories, an ill-balanced diet, a defective protein ration and a large fluid intake. No one of these factors alone seems adequate to produce hypoproteinemia, at least with any constancy.

## FROM THE AUTHOR'S DISCUSSION.

RELATION OF MICROLITHIS TO BILIARY CONCRETIONS. G. LEMMEL AND W. BÜTTNER, Beitr. z. path. Anat. u. z. allg. Path. **92**:262, 1933.

Gallbladder bile containing microliths, obtained at necropsy and at operation, was studied microscopically to determine whether microliths enter into the formation of macroscopic concretions. Growth of microliths by accretion, sufficient to form macroscopic concretions, was not observed, but the union of two or more microliths in a uniting ground substance may lead to the formation of structures visible to the eye. Such masses may form the centers of pigment-calcium calculi.

O. T. SCHULTZ.

SPECTROGRAPHIC DETERMINATION OF COPPER IN TISSUES. W. GERLACH AND K. RUTHARDT, Beitr. z. path. Anat. u. z. allg. Path. **92**:347, 1933.

In this contribution to Gerlach's series of studies on the spectrographic elementary analysis of tissues, a method is given for the detection and quantitative estimation of copper. The advantages of the method, as compared with the electrolytic procedure, are that it may be used on either fresh or fixed tissues, that it is rapid, and that only from 0.3 to 0.6 Gm. of formaldehyde-fixed tissue is necessary for an analysis.

O. T. SCHULTZ.

PHYSICAL AND CHEMICAL CHANGES IN THE SERUM OF INFECTED ANIMALS.  
E. REMY, Ztschr. f. Immunitätsforsch. u. exper. Therap. **81**:57, 1934.

In guinea-pigs with tuberculosis, the examination of various physical properties showed no essential deviations from the normal; in guinea-pigs with trypanosomiasis, a slight lowering of surface tension was found. Tuberculous guinea-pigs showed a marked hyperglycemia and lower values for copper in the blood serum. The serum of guinea-pigs with trypanosomiasis had an increased reducing ability when tested with dichlorphenolindophenol. Syphilitic rabbits showed rises in non-protein nitrogen and in dextrose.

I. DAVIDSOHN.

THE SUGAR CONTENT OF CEREBROSPINAL FLUID IN MENINGITIS. A. FLAUM,  
Acta path. et microbiol. Scandinav., supp. 16, p. 77, 1933.

In cases of bacterial meningitis the percentage of sugar in the cerebrospinal fluid is decreased. In meningitis caused by a "virus" the sugar content is not reduced. The decomposition of the sugar is not due to the cells in the fluid. The sugar may be fermented by bacteria; the virus of poliomyelitis does not break up sugar in the fluid.

JACOB KLEIN.

PATHOGENESIS OF SUBCUTANEOUS ADIPONECROSIS (SCLERODERMIA NEONATORUM). STURE A. SIWE, Acta path. et microbiol. Scandinav., supp. 16, p. 438, 1933.

Areas of subcutaneous infiltration may occur in infants on the shoulders, back, buttocks, cheeks and thighs. They disappear spontaneously leaving behind an atrophic skin associated with cyst formation and calcium deposits. The crystals in the subcutaneous tissue are composed of neutral fats, cholesterol esters and palmitic acid. In animals similar changes may be induced by the injection of palmitin and its acids. In the new-born there is a comparatively high content of palmitic acids.

JACOB KLEIN.

### Microbiology and Parasitology

ACTINOMYCOSIS OF TUBES AND OVARIES. V. H. CORNELL, Am. J. Path. **10**: 519, 1934.

Seventy-one published cases of actinomycosis of the internal female genitalia are listed. Forty-five of the patients died, eight were improved, in seven the outcome was doubtful, and only eleven were possibly cured. The case reported showed involvement of both tubes and ovaries. The patient was operated on and treated by potassium iodide, and is well four years after operation. A tabulation of some features of the published cases is presented.

FROM THE AUTHOR'S SUMMARY.

THE INFECTION OF FERRETS WITH THE VIRUS OF SWINE INFLUENZA. R. E. SHOPE, J. Exper. Med. **60**:49, 1934.

The experiments described confirm the earlier observation of Smith, Andrews and Laidlaw that the virus of swine influenza is pathogenic for ferrets when administered intranasally. A disease that is clinically more severe and pathologically more extensive than that described by Smith, Andrews and Laidlaw is obtained if, when the virus is inoculated, the ferrets are under ether anesthesia. This disease may terminate fatally. Ferrets infected in this way show at autopsy an edematous type of pneumonia of lobar distribution. When stored in 50 per cent glycerol at refrigeration temperature the virus maintains its pathogenicity for ferrets for as long as seventy-five days. After serial passage through sixteen ferrets the virus is still

capable of inducing swine influenza when mixed with a culture of *Haemophilus influenzae-suis* and administered intranasally. Passage through ferrets causes no apparent attenuation of the virus for swine. Serum from pigs recovered from swine influenza is capable of neutralizing the ferret-passed virus for either swine or ferrets. Likewise serum from recovered ferrets neutralizes the virus of swine influenza for either ferrets or swine.

FROM THE AUTHOR'S SUMMARY.

THE RESPIRATION MECHANISM OF PNEUMOCOCCUS. M. G. SEVAG and L. MAIWEG, *J. Exper. Med.* **60**:95, 1934.

A virulent pneumococcus on being transformed into its avirulent form consumes many times more oxygen than the parent organism; but this gain of activity is a temporary property. After a time the pneumococcus degenerates into a form which consumes very much less oxygen than either the virulent or the recently derived avirulent form. These phenomena should receive consideration in any comparative study of the metabolic functions and oxidation products of virulent and avirulent strains of pneumococci. The change that takes place in the structure of the enzyme responsible for the carbohydrate biosynthesis during the shift from the virulent to the avirulent form may be associated with the changes in the enzyme structure already demonstrated in connection with these metabolic studies.

FROM THE AUTHORS' CONCLUSIONS.

SPONTANEOUS CONJUNCTIVAL FOLLICULOSIS OF RABBITS. P. K. OLITSKY, J. T. SYVERTON and J. R. TYLER, *J. Exper. Med.* **60**:107, 1934.

Spontaneous conjunctival folliculosis is widespread among various species of rabbits. It exists in two forms: type 1, in which the lesions are localized and the disease is relatively inactive, and type 2, in which the follicles are closely distributed over the entire surface of the conjunctiva and the process is more active and characterized by extensive inflammatory reactions. One type can be converted into the other either by experimental methods or by natural processes. The disease can be transmitted from rabbit to rabbit by means of subconjunctival inoculation of suspensions of the affected tissues or by instillation of such material into the conjunctival sac, or even by mere contact of folliculotic animals with those having smooth conjunctivae. It is plain that the disease is an infection. The causal agent is not filtrable through Seitz disks that retain *Serratia marcescens* nor through Berkefeld V candles that permit the passage of this organism. Furthermore, the lesions of the spontaneous or of the experimental disease do not exhibit the cytotropic effects or the inclusion bodies suggestive of the action of an ultra-microscopic virus. On the other hand, the lesions are characterized by a persistent, progressive chronicity and show certain resemblances to the granulomas. The evidence suggests that the spontaneous conjunctival folliculosis of rabbits is due to a micro-organism—one having a low grade pathogenic action. In a paper shortly to be published, a bacterium capable of reproducing folliculosis in normal rabbits will be described.

FROM THE AUTHORS' CONCLUSIONS.

VARYING INFLUENCE OF TUBERCULOUS RABBIT PLASMA ON THE GROWTH OF FIBROBLASTS IN VITRO. H. F. SWIFT, J. K. MOEN and E. VAUBEL, *J. Exper. Med.* **60**:149, 1934.

Plasma obtained from tuberculous rabbits within three or four months following their inoculation with bovine tubercle bacilli exerted a growth-inhibitory influence on transplants of rabbit fibroblasts, while that obtained after the fourth month was growth-stimulating. It is suggested that the inhibitory factor was linked in part with lipoidemia, while the stimulating elements were associated with leukocytosis.

FROM THE AUTHOR'S SUMMARY.

**THE FATE OF BCG AND ASSOCIATED CHANGES IN THE ORGANS OF RABBITS.**  
M. B. LURIE, J. Exper. Med. 60:163, 1934.

Under the conditions of experiments in which 1 mg. of BCG is introduced intravenously into rabbits, the BCG multiply in the body, but they are soon destroyed, completely in most organs, all but completely in the lymph nodes. The remaining BCG persist in the lymph nodes for a long time, causing no tuberculous changes, and acquiring no added virulence for rabbits. The BCG produce typically tuberculous changes, sometimes extensive, which resolve completely. They cause caseation, but no softening. Acquired local immunity in tracheobronchial, mesenteric and axillary lymph nodes and in the spleen is shown to be less effective for a time than that in other organs. The destruction of bacilli begins with the appearance of cutaneous sensitivity to tuberculin and is at its height with maximum sensitivity. The secondary acute inflammatory reactions in and about tuberculous foci, the caseation and the hypersensitivity to tuberculin develop synchronously. Caseation and sensitivity to tuberculin do not occur early in the course of the disease in response to considerable amounts of bacilli and of tuberculin, but later they are incited by smaller amounts.

FROM THE AUTHOR'S CONCLUSIONS.

**RESULTS OF IRRADIATING STAPHYLOCOCCUS AUREUS BACTERIOPHAGE WITH MONOCHROMATIC ULTRAVIOLET LIGHT.** F. L. GATES, J. Exper. Med. 60: 179, 1934.

The incident energies required to kill Staphylococcus aureus and to inactivate its homologous bacteriophage have been measured at the various wavelengths of the quartz mercury vapor arc between 238 and 302 millimicrons and found to run strictly parallel, the readings for the Staph. aureus phage being obtained at a uniformly higher level of energy. This difference in levels is of less significance than the striking similarity in the shapes of the energy curves, which indicate that in both instances the same organic structures are absorbing the radiations. The results are open to three interpretations. The most obvious is that the bacteriophage is a submicroscopic organism. Again, it is possible that the bacteriophage is a product of its own lytic action on the homologous bacterium and contains the essential structural units which in Staph. aureus also are destroyed by ultraviolet rays, causing the death of the organism. A third, more remote explanation is that the phage, of wholly unknown nature, is absorbed on Staph. aureus material in so intimate a bond that the alteration of this material by irradiation renders the phage incapable of further lytic activity.

FROM THE AUTHOR'S SUMMARY.

**THE FIBRINOLYTIC ACTIVITY OF HEMOLYTIC STREPTOCOCCI.** R. L. GARNER and W. S. TILLETT, J. Exper. Med. 60:239 and 255, 1934.

The active fibrinolytic principle in cultures of hemolytic streptococci can be isolated in stable form and partially purified by the following methods: (1) precipitation of the culture filtrate with 3 volumes of 95 per cent ethyl alcohol; (2) adsorption on especially prepared aluminum hydroxide according to Willstätter followed by elution with tenth-molar sodium phosphate buffer, *pH* 7.3. Concentration can be accomplished best by vacuum dialysis of either the culture filtrates or the preparations obtained by adsorption and elution. The streptococcal fibrinolysin is characterized by the following properties: 1. It may resist heating to 100 C. for sixty minutes; variations in thermal resistance are described. 2. Partially purified preparations give positive reactions in tests for protein. Activity is rapidly and completely destroyed by trypsin or papain. The active principle is demonstrable in dissolved fibrin even after eighteen hours' incubation.

The fibrinolysin of hemolytic streptococci exerts no hydrolytic action on casein, gelatin or peptone. The action on solid human fibrin is characterized by a small and gradual increase in the aminonitrogen content of the solution. The specific and special enzymatic action of fibrinolysin is contrasted with the actions

of trypsin and streptococcic peptase. Solutions of human fibrinogen, after brief incubation with fibrinolysin, lose the capacity to form fibrin. Solutions of rabbit fibrinogen, on the other hand, retain the property of transformation into fibrin even after prolonged exposure to fibrinolysin. Qualitative tests with solutions resulting from the action of streptococcic fibrinolysin on human fibrin indicate that the end-product may be protein and that the degradation of the molecule is not great.

## FROM THE AUTHORS' SUMMARIES.

PULMONARY INFECTION IN PNEUMOCONIOSIS. H. O. PROSKE and R. R. SAYERS, Pub. Health Rep. 49:839, 1934.

In general, the silicotic lung is more susceptible to bacterial infection than the average lung. This is probably due to the irritation of the respiratory tissues by the inhaled dust particles which weakens the mucous membranes and renders them susceptible to infection. The toxic influence of certain inorganic dusts on the tissues may be a contributing factor. The relation of tuberculosis to pneumoconiosis has been studied to a considerable extent, but comparatively little work has been done in connection with other infectious processes of the lung, e. g., pneumonia, pulmonary abscess, bronchiectasis and influenza. An investigation was made of these conditions, both bacteriologically and experimentally, with the view of obtaining a better understanding of the predisposition to, and the mechanism of, infection of the lung in certain dusty trades. Bronchiectasis, abscess of the lung and gangrene occur frequently in hard-rock miners. It has been definitely established that aerobic pathogenic bacteria and certain fungi are responsible for these conditions, but the high percentage of cases in which the anaerobic microbes of the mouth and throat have been reported suggests that they at least participate in the production of the diseases. The responsibility of fusospirochetal organisms for severe infections of the respiratory tract had been suspected as early as 1867. In the past few years more than 2,000 cases of fusospirochetal abscess of the lung have been reported in the United States. Accurate figures on bronchiectasis of the same origin are not available. The mode of infection of the lungs in persons not engaged in dusty trades is briefly discussed and compared with the possible mechanism of infection in those having pneumoconiosis. The bacteriology of fusospirochetal disease of the lung is given in detail, and a practical technic for the study of the anaerobic flora of the upper respiratory tract is appended.

## FROM THE AUTHORS' SUMMARY.

BACTERIOLOGY OF THE TUBERCULOUS PRIMARY COMPLEX AT VARIOUS AGES. H. E. ANDERS, Beitr. z. Klin. d. Tuberk. 81:260, 1932.

The question of the length of time that viable tubercle bacilli persist in calcified foci in lymph nodes has great biologic and clinical importance. Bibliographic research reveals no general agreement on this point. Bacteriologic studies on 157 anatomically healed primary foci have been reported. Tubercle bacilli were cultured from the lymph node component 36 times. Most of the positive cultures were obtained from persons above the fifth decade of life, and the negative cultures from those below the fifth decade. Few bacteriologic studies have been reported for intestinal primary complexes. Iizulia studied 39 such foci histologically without finding any bacilli. Anders studied 32 isolated primary pulmonary complexes and found viable bacilli in 6, an incidence of 21.4 per cent. All of these were from persons in the fifth or sixth decade of life. The negative ones were from persons between 20 and 40 years of age. No bacilli could be cultured from 14 healed intestinal foci. The conclusions are: that viable bacilli probably persist longest in those infected relatively late in life; that the source of endogenous lymphoglandular reinfection is not invariably the lymph node component of the primary complex as previously thought, but also, probably, lymph nodes in the same drainage area which show no histologic tuberculosis but contain viable bacilli, and finally that intestinal primary foci are probably only rarely the source of such endogenous reinfection.

AARON EDWIN MARGULIS.

**EXPERIMENTAL SYPHILIS OF THE CENTRAL NERVOUS SYSTEM.** T. TANI and H. FUNADA, Zentralbl. f. Bakt. (Abt. 1) **125**:423, 1932.

Rabbits were inoculated intracerebrally with testicular emulsions of four strains of spirochetes, three obtained from condylomas and one from a patient with yaws. Each strain initiated infection, as shown by the development of a positive Wassermann reaction of the spinal fluid. Tani and Funada conclude that this speaks against the idea of neurotropic strains of spirochetes. Furthermore, fourteen rabbits into which spirochetes from rat-bite fever, or Trypanosoma gambiense, were injected intracerebrally yielded positive Wassermann reactions of the blood but negative reactions of the spinal fluid. The conclusion, therefore, is that the antigenic substance which engenders the Wassermann reagent comes from the treponema.

Rabbits which had been infected with syphilis either in the testicle or the brain several months previously, and whose somatic organs were resistant to reinfection, were inoculated intracerebrally with spirochetal testicular emulsions. The spinal fluid in these animals became Wassermann-positive. The authors take this to mean that the central nervous system of rabbits is less resistant to syphilitic reinfection than are the somatic organs, rather than that there is a neurotropism of spirochetal strains.

PAUL R. CANNON.

**THE EFFECT OF PHYSIOLOGIC SOLUTION OF SODIUM CHLORIDE ON STAPHYLOCOCCUS AUREUS.** A. LAURELL, Acta path. et microbiol. Scandinav. supp. 16, 1933, p. 204.

Washing staphylococci with physiologic solution of sodium chloride had two toxic effects: (1) a bactericidal action due to oligodynamic influences in distilled water and (2) a detoxicating action, attributed to the sodium chloride ion, whereby the staphylococci become nonpathogenic in animals. No difference in agglutination titer was observed between staphylococci detoxicated in salt solution and those treated with formaldehyde.

JACOB KLEIN.

**RESORPTION OF BACTERIA FROM THE GASTRO-INTESTINAL TRACT.** ARVID LINDEN, Acta path. et microbiol. Scandinav., supp. 16, 1933, p. 225.

Behring, Ehrlich and others demonstrated that in new-born animals proteins and antitoxins pass unchanged from the gastro-intestinal tract into the blood. There is considerable disagreement about the question of resorption of micro-organisms from the gastro-intestinal tract. Calmette's peroral vaccination against tuberculosis has awakened interest in this question. Linden gave india ink, carmine, timothy bacilli, bovine tubercle bacilli and BCG to young mice and guinea-pigs. Microscopic examination indicated that resorption of these substances and bacteria occurred rarely. The results of the study are not in agreement with Calmette's assumption of a general impregnation of the lymphoid system by the ingested bacteria.

JACOB KLEIN.

### Immunology

**NEMATODE AND CARCINOMA IN HUMAN KIDNEY PELVIS.** ALFRED PLAUT, Am. J. Cancer **20**:610, 1934.

A painful abdominal swelling which had been noticed a few weeks before admission proved on examination to be a large kidney with stones. At operation many cysts were found with firm tissue between them, suggestive of a neoplasm. Six weeks after nephrectomy the patient was operated on again on account of swelling and pain. The broken-down tissue which was removed at this operation proved to be squamous cell carcinoma. The carcinoma, which obviously started from the renal pelvis, was densely infiltrated with eosinophil cells. The remainder of the renal tissue was severely inflamed and widely atrophic.

At autopsy, metastatic squamous cell carcinoma was found in the liver and retroperitoneal tissue. Calcified masses in the kidney were suggestive of remnants of a worm. Continued examination revealed a characteristic but unusually small nematode egg. This egg cannot belong to any of the nematodes which have been described in the kidney so far. The occurrence of parasitism and tumor of the kidney together is regarded as extremely rare in human and animal pathology. It is possible that the presence of the worm was a factor in the development of the carcinoma.

The patient was Russian and had been a resident of New York City for twenty-one years.

#### AUTHOR'S SUMMARY.

**ACQUIRED AND GENETIC IMMUNITY.** J. W. GOWEN AND R. G. SCHOTT, Am. J. Hyg. **18**:674 and 688, 1933.

The data herein presented show that the ability to survive a given inoculation of the virus of pseudorabies is markedly influenced by the genetic constitution of the animal. Susceptibility shows some tendency to be dominant in the  $F_1$  cross. Comparison of a line which was resistant to pseudorabies and another which was susceptible for their respective resistances to another disease, mouse typhoid, showed their reactions to the second disease completely reversed. The  $F_1$  cross for the second disease now show susceptibility largely recessive. These facts lead to the conclusion that genetic constitution as it is related to resistance to these diseases is perhaps best regarded as a composite of several distinct genes, some favoring resistance or susceptibility to one environmental agent and some to another.

Double mating is suggested as a genetic technic for distinguishing between acquired and inherited immunity. This technic seems to favor the hypothesis that resistance to inoculated *Salmonella aertrycke* in a selected strain of mice is due to a concentration of genetic factors for resistance and not to a transfer of acquired passive or active immunity.

#### FROM THE AUTHORS' SUMMARIES.

**A STUDY OF THE GROWTH IN AREA OF INTRACUTANEOUS TUBERCULIN REACTIONS.** C. A. STEWART, Am. Rev. Tuberc. **28**:844, 1933.

The degree of specific allergy to tuberculin induced in a group of children by a primary tuberculous infection varies widely in different ones. The average degree of allergy in children who have primary tuberculosis exclusively is represented by an area of about 30.2 sq. cm. for 0.1 mg. of old tuberculin. The area of the cutaneous reaction to tuberculin is not directly proportional to the amount of tuberculin used. The time required to attain the maximum area is directly related to the degree of allergy, and the relative rapidity with which the impulse to area increment is expended is inversely related to the degree of allergy.

H. J. CORPER.

**THE AGGLUTINATION OF HEMOLYTIC STREPTOCOCCI BY PLASMA AND FIBRINOGEN.** W. S. TILLETT and R. L. GARNER, Bull. Johns Hopkins Hosp. **54**:145, 1934.

Plasma from normal persons and from patients is capable of agglutinating, in very high dilutions, certain strains of hemolytic streptococci. Fibrinogen, chemically isolated from plasma, also agglutinates the same strains of hemolytic streptococci and is considered to be chiefly responsible for the reactivity of the plasma. Only those strains of hemolytic streptococci that are agglutinated by serums from persons harboring acute infection are visibly affected by plasma or fibrinogen; killing the organisms by heat destroys their reactivity in plasma and fibrinogen, as well as in serum; cultures, killed with formaldehyde, remain susceptible to the effect of each of the three blood constituents. The reactive fraction in the serums of patients seems to be a protein closely related to fibrinogen.

#### FROM THE AUTHORS' CONCLUSIONS.

THE RELATION OF ALLERGY TO IMMUNITY IN TUBERULOSIS. H. ROTHSCHILD,  
J. S. FRIEDENWALD and C. BERNSTEIN, Bull. Johns Hopkins Hosp. 54:232,  
1934.

Complete desensitization to tubercle bacilli and to tuberculin can be achieved in tuberculous guinea-pigs by a prolonged and properly graded course of subcutaneous injections of Koch's old tuberculin. The desensitizing power of purified tuberculoprotein is less than that of Koch's old tuberculin in proportion to its lesser power to produce allergic reactions. Long-continued daily subcutaneous injections of massive doses of concentrated glycerin broth in some instances desensitizes tuberculous guinea-pigs to tuberculin. This nonspecific desensitization is not due to the glycerin contained in the broth. It is not known whether this nonspecific desensitization is free from the danger of perifocal reaction. Infection in desensitized immune animals does not introduce into the histologic picture of tuberculous lesions features that are novel to the pathology of human tuberculosis. In all these respects the reactions of the desensitized animals were equal to or superior to those of the nonsensitized controls. So far as inhibition of the spread of lesions from the site of infection to the viscera may be used as evidence of a local fixation of bacilli, the desensitized, nonallergic immune animals were able to resist the spread of infection as successfully as the allergic ones.

FROM THE AUTHORS' CONCLUSIONS.

A STUDY OF THE CHARACTER AND DEGREE OF PROTECTION AFFORDED BY THE IMMUNE STATE INDEPENDENTLY OF THE LEUCOCYTES. A. R. RICH and C. M. MCKEE, Bull. Johns Hopkins Hosp. 54:277, 1934.

When the circulating leukocytes are sufficiently reduced by benzene none emigrate at a site of infection, and the character of the protection afforded by an immune state, independently of that conferred by the activity of the leukocytes, can be studied. Immunized animals were treated with benzene to remove their leukocytes, and were then infected intradermally with virulent pneumococci of type I. In these immunized leukopenic animals the immune antibody influenced profoundly both the character of the bacterial growth and movement and the course of the infection. The bacteria, as they proliferated, adhere to themselves and apparently to the tissues as well, and were thus held fixed at the site of inoculation for hours after nonimmune controls had died of septicemia. The immediate local immobilization of the bacteria was demonstrable even in the absence of microscopically detectable inflammatory exudate or thrombosis of lymphatics. The latter factors are, therefore, not primarily responsible for the immobilization of the bacteria in the immune body, though when they are finally established they undoubtedly serve to assist in inhibiting the spread of the bacteria. Since the phenomenon of immobilization occurs in passively as well as in actively immunized animals, it is the antibody content of the fluids of the immune body which is primarily responsible for this prevention of the prompt spread of the bacteria. However, in the absence of the leukocytes the growth of the immobilized bacteria proceeds uninterruptedly until great colonies and masses of pneumococci have been formed at the site of infection, after which the bacteria penetrate into the blood and lymph streams and the animal dies with septicemia even though its plasma is potent in passively protecting nonimmune animals possessing leukocytes. If only relatively few leukocytes appear at the site of infection (far fewer than the number which appear in the lesions of nonimmune controls which succumb to the infection) the bacteria, which are opsonized by the antibody, are rapidly ingested and destroyed, and the lesion is sterilized. In addition to its opsonizing power, the humoral antibody, therefore, performs the important protective function of preventing the immediate spread of the bacteria throughout the body, holding them fixed at the site where they lodge until the phagocytic leukocytes are able to reach the spot and destroy them. It is, furthermore, the humoral antibody which, with the cooperation of the intravascular phagocytic macrophages of the liver and spleen,

brings about the rapid segregation and destruction of bacteria which do penetrate into the blood stream, thus further inhibiting the development of septicemia and metastatic infection; but acquired immunity creates no condition of the fluids or fixed tissues which can prevent the progressive and overwhelming growth of the bacteria in the absence of the phagocytes. **FROM THE AUTHORS' SUMMARY.**

**PERIVASCULAR REACTIONS FOLLOWING INJECTION OF STREPTOCOCCI INTO SENSITIZED RABBITS.** C. H. HITCHCOCK and others, *J. Exper. Med.* **59**:283, 1934.

Intravenous injection of small doses of nonhemolytic streptococci into previously sensitized rabbits is usually followed by the appearance of perivascular cellular aggregates in the lungs and liver. The characteristic cell in these aggregates is moderately large, with a vesicular nucleus, prominent nucleoli, clumped chromatin and basophilic cytoplasm. In addition, the lesions contain small lymphocytes and granulocytes. This lesion is easily differentiated by architecture and cell content from normally occurring lymphoid aggregates and from spontaneous rabbit hepatic cirrhosis. This mononuclear response does not occur when the intravenous dose is large enough to cause death of the animal within twenty-four hours. In the spleen and lymph nodes the characteristic basophilic cells which normally occur in these organs are present in increased numbers. Following intravenous treatment alone or sensitization without intravenous treatment, the lesions occur much less frequently and, when present, are smaller and more sparsely found. As, in the present series of experiments, this lesion was not found in normal animals and infrequently in those treated by the intravenous route alone, it is suggested that the preliminary sensitization serves to enhance the animal's reactivity to the antigen. In this way a small dose of bacteria is capable of eliciting the cellular phenomenon, which in unsensitized animals appears only when larger doses of antigen are administered over longer periods of time. Too large a dose of antigen, however, results in shock and cell death rather than in proliferation.

**FROM THE AUTHORS' SUMMARY AND CONCLUSIONS.**

**A SEROLOGICAL DIFFERENTIATION OF BOVINE HEMOLYTIC STREPTOCOCCI (GROUP B).** R. C. LANCEFIELD, *J. Exper. Med.* **59**:441, 1934.

Hemolytic streptococci of group B (derived chiefly from cattle) have been further subdivided by use of the precipitin reaction into specific types. With three exceptions, the twenty-one strains of group B were differentiated into three specific types. Chemical analyses of the type-specific substances of group B strains of types I and II show that they are polysaccharides (S substances). This is in contrast to the fact that proteins (M substances) were previously shown to determine type specificity among strains of human origin (group A). The group-specific substance, C, serologically identical in all members of group B, was also identified as of polysaccharide nature.

**FROM THE AUTHOR'S SUMMARY.**

**LOSS OF HEMOLYSIN AND PIGMENT FORMATION WITHOUT CHANGE IN IMMUNOLOGICAL SPECIFICITY IN A STRAIN OF STREPTOCOCCUS HAEMOLYTICUS.** R. C. LANCEFIELD, *J. Exper. Med.* **59**:459, 1934.

A variant arising in a culture of a hemolytic streptococcus was shown to have lost the properties of producing pigment and hemolyzing blood. Despite the loss of these two functions, it had in common with the strain from which it was derived certain other distinguishing biochemical characteristics, as follows: Both attained the same hydrogen ion concentration in dextrose broth; both hydrolyzed sodium hippurate, grew on bile agar, and fermented trehalose but not sorbitol; both failed to reduce methylthionine chloride U. S. P. (methylene blue) in milk cultures and were insusceptible to the action of streptococcal bacteriophage. In addition, the

virulence of the variant remained the same as that of the original culture. The antigenic and serologic specificity of the variant was identical with the group and type specificity of the original strain (group B, type I). These specificities were established by the use of immune serums prepared by immunization of rabbits with each form. The immunologic reactions employed were reciprocal agglutination, precipitation, agglutinin absorption, precipitin absorption and passive mouse protection.

FROM THE AUTHOR'S SUMMARY.

STUDIES ON TYPHUS FEVER. H. ZINSSER and M. R. CASTANEDA, *J. Exper. Med.* **59**:471, 1934.

Guinea-pigs infected with European typhus virus can be passively protected with the serum of a horse that has been treated with killed Mexican rickettsiae.

FROM THE AUTHORS' CONCLUSIONS.

ON THE USE OF ADSORBENTS IN IMMUNIZATIONS WITH HAPTONS. J. JACOBS, *J. Exper. Med.* **59**:479, 1934.

Experiments are described which confirm the observation of Gonzalez and Armangué that heterogenic extracts can be made antigenic by adsorption to inorganic materials. With fractions of the original extracts from which a part of the inactive material had been removed no such enhancement was observed, whereas with foreign protein an activation was still possible. Carbohydrate preparations behaved similarly in that purification, perhaps loss of protein, was accompanied by a distinct decrease in antigenicity after adsorption. The activity of a but slightly antigenic hetero-albumose preparation was markedly increased after adsorption to charcoal and alum. The most reasonable explanation for the effects observed by Gonzalez and Armangué, and Zozaya, seems to be that a preexisting antigenic capacity has been enhanced by the use of adsorbents. The experiments reported here support the view that these effects are influenced significantly by the presence of substances other than those of a specific nature.

FROM THE AUTHOR'S SUMMARY.

HETEROPHILE ANTIGEN COMMON TO AVIAN ERYTHROCYTES AND SOME STRAINS OF PASTEURELLA. L. BUCHBINDER, *J. Immunol.* **26**:215, 1934.

A new heterophile antigen present in the erythrocytes of birds and in some strains of the group of bacteria concerned in hemorrhagic septicemia is described. In contrast to its apparent chance appearance in strains of Pasteurella, it is present in many birds in an orderly fashion. The significance of this heterophile antigen in avian erythrocytes is discussed from the standpoint of the evolution of species. Additional strains of the hemorrhagic septicemia organism containing Forssman's heterophile antigen are described.

FROM THE AUTHOR'S SUMMARY.

PNEUMOCOCCUS LEUCOCIDIN. F. Oram, *J. Immunol.* **26**:233, 1934.

A toxin has been produced from actively growing cultures of Pneumococcus which destroys leukocytes, as demonstrated by the Neisser and Wechberg method. This leukocidin has been demonstrated in aerobic and anaerobic cultures, from growths of both virulent and avirulent strains and from types I, II and III. It is believed that a peroxide which is formed in the aerobic cultures modifies the potency of the toxins through oxidation. The peroxide appears earlier and is of greater concentration in cultures of the virulent strains; this may account for the avirulent cultures apparently containing toxins of as great a potency as those of the virulent strains.

FROM THE AUTHOR'S SUMMARY.

EFFECT OF IMMUNE YELLOW FEVER SERUM IN MONKEYS. N. C. DAVIS, J. Immunol. **26**:361, 1934. PROTECTIVE PROPERTIES AGAINST YELLOW FEVER VIRUS IN THE SERA OF THE OFFSPRING OF IMMUNE RHESUS MONKEYS. M. HOSKINS, *ibid.*, p. 391.

Immune serum from recovered animals, when injected at from twenty-four to forty-eight hours following inoculation with the virus of yellow fever, was capable of preventing the fever or ameliorating it in a significant proportion of experimental monkeys. After forty-eight hours the effect was less definite. In no instance did the serum prevent death when administration was delayed until the temperature of the monkey had reached 104 F. (Davis.)

Five baby monkeys born of mothers immune to yellow fever were all found to have in their serum protective properties against yellow fever. This was the case when they were still subsisting on their mother's milk. In two instances in which the baby monkeys had been separated from their mothers for three weeks, the serum of the offspring showed no evidence of protective properties. (Hoskins.)

FROM THE AUTHORS' SUMMARIES.

IMMUNOLOGIC STUDIES IN TYPHOID FEVER WITH RELAPSES. G. D. C. THOMPSON and E. E. ECKER, *J. Infect. Dis.* **54**:177, 1934.

A case of typhoid fever with two relapses is reported which is of particular interest because of a complete lack of agglutination and precipitation reactions in the course of the disease while positive complement fixation and bacteriolytic and marked opsonic powers were observed, possibly indicating the final mechanisms of recovery. These results also appear to indicate a plurality of antibodies, but it is to be emphasized that the antigen is complex and that the question of unity or plurality of antibodies can be solved only by the use of a single and pure antigen. The case is also of importance in that the serum failed to dissociate the organism and because no bacteriophage was isolated. The organisms isolated proved to be typical typhoid bacilli (culturally and serologically) and stimulated the production of the O type of agglutinins in the rabbit.

FROM THE AUTHORS' SUMMARY.

GRANULAR AND FLOCCULAR TYPES OF AGGLUTINATION WITH TYPHOID BACILLUS. R. GILBERT, M. COLEMAN and A. B. LAVIANO, *J. Lab. & Clin. Med.* **19**:225, 1933.

The employment of macroscopic tests with two suspensions of killed typhoid bacilli, one to demonstrate the floccular or species-specific, and the other, the granular or group-agglutinative properties, usually furnishes information of greater diagnostic significance than the microscopic test with a living culture does. Apparently, agglutination in a 1:80 or higher dilution with an alcohol-treated suspension usually indicates that the patient has typhoid fever or an infection incited by a species allied to *Bacillus typhosus*, while a similar reaction with a formaldehyde-treated suspension suggests one of three alternatives, that the patient has typhoid fever, that he has had the disease in the past or that he has received typhoid vaccine. Both the granular and the floccular types of agglutination have seldom been observed in high dilutions of serum other than that from patients with typhoid fever.

FROM THE AUTHORS' SUMMARY AND CONCLUSIONS.

ATTEMPTS TO LOCATE THE SITE OF ANTIBODY PRODUCTION. G. A. H. BUTTLE, *Brit. J. Exper. Path.* **15**:64, 1934.

Some have thought that the reticulo-endothelial system is the site of the production of antibodies; others have been of the opinion that all tissues are involved. It has been reported that tissue cultures produce antibodies. The author injected into rabbits a 2 per cent alum-precipitated diphtheria toxoid and after a month

titrated the antitoxic power of the blood. Then an exsanguination and transfusion were done so that four fifths of the circulating blood was substituted by normal rabbit blood. It was found that the blood does not produce antitoxin, but acts merely as a vehicle in which it is carried around the body after it is formed by the tissues. Removal of the liver, spleen and skin did not affect the rate of antitoxin production in rabbits. Probably all the tissues take part in the production of antibodies.

JACOB KLEIN.

ABSORPTION OF ANTITOXIN THROUGH THE SKIN. R. RICHOU, Ann. Inst. Pasteur 51:117, 1933.

Tetanus antitoxin in various forms was applied to the skin of guinea-pigs. Percutaneous absorption was manifested, but the skin appeared to receive antitoxin only passively, permitting some slight general absorption. Only about 1 part in 1,000 had any effect. "Between the effect of antitoxin introduced by one or the other of these ways there was no qualitative difference, but only quantitative differences, the disadvantage remaining, from many points of view, with the percutaneous method."

M. S. MARSHALL.

PERCUTANEOUS SERUM SENSITIZATION. R. RICHOU, Ann. Inst. Pasteur 51:146, 1933.

Following percutaneous application of tetanus antitoxin guinea-pigs were found to be sensitized to serum to an extent permitting anaphylactic shock. Such sensitized animals were not desensitized by percutaneous application of serum a day before the test shock dose was given.

M. S. MARSHALL.

RED BLOOD CELLS AND IMMUNITY. R. DUJARRIC DE LA RIVIÈRE and N. Kossovitch, Ann. Inst. Pasteur 51:149, 1933.

Red blood cells adsorb diphtheria toxin; the adsorption capacity of the cells varies with the species of animal; cell stroma fails to fix toxin or fixes it in minimum quantity. Tetanus anatoxin fixed on red blood cells preserves them from the action of the corresponding toxin. The serum of animals immunized with a hemoglobin which has been submitted to several successive crystallizations possesses antibodies which corresponded to the antigen and which are rigorously specific.

AUTHORS' CONCLUSIONS.

ADENOCARCINOMA AND SQUAMOUS CELL CARCINOMA. J. NOCHIMOWSKI, Frankfurt. Ztschr. f. Path. 44:547, 1933.

This is a review of the literature and a report on four additional cases. As to the pathogenesis of these tumors it appears that two possibilities must be considered. One is that both the adenocarcinoma and the squamous cell carcinoma, arising from different parts, are growing into each other and thus give rise to the formation of "collision tumor." It is also possible that such tumors may arise from cells of so little differentiation that they are capable of developing into both adenocarcinoma and squamous cell carcinoma. These rare tumors are classified by Nochimowski as "true" or "primary" adenocarcinoma and squamous cell carcinoma.

WILLIAM SAPHIR.

PECULIAR TUMOR ARISING IN THE GONADS OF AN INTERSEX. E. BRAUER, Frankfurt. Ztschr. f. Path. 45:224, 1933.

An 18 year old person revealed a relatively small penis with hypospadias and absence of the prostate, seminal vesicles and testes. Because of a sudden attack of pain in the lower quadrant and accompanying fever, the patient was operated

on. A uterus and two tubes were found, and in place of the ovaries two large tumors, one of which appeared twisted and necrotic. The tumors and both tubes were removed. The patient died subsequently, and permission for a postmortem examination could not be obtained. Each tumor weighed over 3 pounds (1.4 Kg.). They were yellowish white and contained coarse nodules. The tubes histologically corresponded to those seen in females about 12 years old. The tumors consisted of connective tissue septums, between which heaps of cells were recognizable, with prominent nuclei. The cells were situated very close to one another and were surrounded by connective tissue which formed alveolus-like spaces. No blood vessels were seen within the tumors. The tumor cells were occasionally arranged in rows and islets, and sometimes presented a fanlike appearance. In some fields the connective tissue stroma and in others the epithelial elements predominated. This was so marked that Brauer concluded that the tumor was either an alveolar sarcoma or a carcinoma. Only a few lumens were seen, lined by large cuboidal epithelial cells.

O. SAPHIR.

**GRANULOSA CELL TUMORS OF THE OVARIES.** H. BETTINGER, Frankfurt. Ztschr. f. Path. 45:238, 1933.

The first instance concerns a 56 year old woman who had suffered from irregular metrorrhagias. Examination of the specimen obtained by curettage led to a diagnosis of adenocarcinoma of the fundus. A panhysterectomy revealed a carcinoma in addition to two myofibromas; also that the left ovary was cystic and that the right showed many firm nodular tumors. The latter consisted of connective tissue fibers rich in nuclei with many intercalated fibers. Many large nests of cells were found within the stroma. The cells were polyhedral, containing small nuclei with much chromatin and a lightly stained cytoplasm. Only occasionally a slight tendency toward the formation of cysts was observed in the centers of the cellular foci. A second case was found in a 57 year old woman, who had a typical pseudomucinous cystoma in the left ovary. The right ovary contained a tumor similar to the one described in the ovary in the preceding case.

A second type of tumor was found in a 17 year old girl who had broad shoulders, small hips and absence of pubic hairs. The mammary glands were underdeveloped, and menstruation had not occurred. At operation, an infantile uterus was found, and a large solid tumor of the left ovary and a small tumor of the right were present. These tumors consisted of a connective tissue stroma with many lymphocytes; many foci of large cells with large round nuclei and much cytoplasm. No particular arrangement of the cells was noticeable. A sister of this patient, who had shown a similar tumor of the ovary, died two years after the operation as a result of generalized metastases.

O. SAPHIR.

**RETICULAR LYMPHO-EPIHELIOMA OF THE THYMUS.** E. LAAS, Frankfurt. Ztschr. f. Path. 45:309, 1933.

An instance is described in a 62 year old man. The tumor was found beneath the sternum at the site of the thymus. It was well circumscribed within the fatty tissue and weighed 55 Gm. There were many spindle-shaped cells and lymphocytes situated within a network of reticulum fibers. Within the reticulum, mesenchymal cells but no tumor cells were present.

O. SAPHIR.

**IMMATURE LOCALIZED, AND DIFFUSELY INFILTRATING RHABDOMYOBlastoma.** M. GLASUNOW, Frankfurt. Ztschr. f. Path. 45:328, 1933.

Six instances of rhabdomyoblastoma are described. Glasunow distinguishes between myoblastomyoma, which is a benign tumor, and myoblastosarcoma or diffusely infiltrating rhabdomyoblastoma, which are of the malignant variety. The so-called myoblastomyomas are well circumscribed, but not encapsulated, tumors.

They consist of round, oval or elongated cells measuring 15 to 30 microns in diameter. They reveal a slightly basophilic and finely granular cytoplasm. Also, syncytial masses are present, often arranged in the form of bundles. The stroma consists of thin collagen fibrils. Occasionally, the tumor elements may be very large. The malignant type is characterized by pleomorphism of cells. Giant cells are often encountered, and mitotic figures are present. The cytoplasm is distinct and finely granular. The finely granular cytoplasm is characteristic of the myoblastoma. Oxyphilic granules are occasionally found in the cytoplasm. A striation of the cytoplasm of the fibers may be encountered. The longitudinal striation is, as a rule, much more pronounced than the cross striation. There is no principal difference between the malignant myoblastoma and the rhabdomyoma.

O. SAPHIR.

MORPHOLOGY OF FAT IN CARCINOMA. B. KELLNER, Frankfurt. Ztschr. f. Path. 45:383, 1933.

The fat content of fifty carcinomas was studied. Fat was found in the tumor cells and in the connective tissue. Squamous cell carcinoma, particularly the type which does not show keratinization, contained the least. Fat probably is carried to the tumor cells by the lymphatics or may be liberated by necrotic cells, but cannot be utilized by the neoplastic cells.

O. SAPHIR.

IMMUNE BIOLOGIC REACTION OF THE TRANSPLANTABLE MOUSE CARCINOMA.  
B. M. MÜHLENBEIN, Frankfurt. Ztschr. f. Path. 45:514, 1933.

Mühlenbein succeeded in producing true anaphylactic reactions in white mice with hog serum. The animals were sensitized with hog serum (0.3 cc.) given at intervals of from three to seven days. Twenty-eight days after the last sensitizing injection, an intravenous administration of the serum was usually followed by death due to anaphylaxis. This method was utilized in an attempt to sensitize white mice with autogenous aqueous tumor extracts. While a true anaphylactic condition could not be brought about, it was found, nevertheless, that the sensitized animals showed a marked resistance against transplantation of the autogenous tumor. In cases of successful transplantation there was a definite retardation of the growth of the tumor.

W. SAPHIR.

IMMUNITY TO THE TRANSPLANTABLE MOUSE TUMOR. G. FISCHER, Frankfurt. Ztschr. f. Path. 45:526, 1933.

Fischer attempted to produce immunity in white mice against transplantable tumors by preceding injections of human tumor material. Aqueous extracts of such material injected repeatedly into white mice did not produce an anaphylactic condition in the animals. The "taking" of transplantable mouse tumor in the treated did not differ from that in the control animals. Likewise, attempts to produce immunity by injections of the globulin fraction and the phosphatid fraction of human tumor extracts gave repeatedly negative results.

W. SAPHIR.

TUMOR GROWTH IN UNDESCENDED AND COMPENSATORY HYPERTROPHIC TESTICLES. E. DE BARY, Frankfurt. Ztschr. f. Path. 45:556, 1933.

De Bary briefly mentions the theories of tumor origin and growth and includes that of Fischer-Wasels, who explains the origin and growth of tumors on the basis of a disturbance either of embryonal development or of regeneration and on Askanazy's "vegetative-activating" factor. According to Fischer-Wasels the anlage of a tumor arises only through a disturbance of embryonal or regenerative development (embryonal heteroplasia and regenerative metaplasia included). Askanazy maintains, however, that such an anlage might follow on a compensatory hyper-

trophic process. De Bary is evidently opposed to the latter view because, notwithstanding the frequency of compensatory hypertrophy of the heart, tumors of that organ are rather rare. Further, a kidney that has undergone compensatory hypertrophy shows a tendency toward inflammatory and degenerative changes rather than toward the formation of a tumor. In order to clarify the issue further he has investigated the incidence of neoplastic growth in undescended and compensatory hypertrophic testicles. A survey of the literature revealed only one questionable case of tumor in the normally placed partner of a cryptorchid testicle. On the other hand, he found, in accord with the literature, a far greater tendency toward the development of tumor in the undescended organ than in the scrotal hypertrophic organ. About 12 per cent of all undescended testicles reveal tumor formation. This tendency to the development of tumor is attributed to a disturbance in embryonal development.

H. HORN.

### Medicolegal Pathology

DEATH FROM DINITROPHENOL. F. E. POOLE AND R. B. HAINING, J. A. M. A. **102**: 1141, 1934. FATAL DINITROPHENOL POISONING. M. L. TAINTER AND D. A. WOOD, J. A. M. A. **102**:1147, 1934.

Poole and Haining describe a case of death following the taking of 2,880 mg. of dinitrophenol in the course of five days by a woman 25 years old. A few hours before death the patient became restless, perspired profusely and complained of "burning up"; the temperature was at least 101.8 F.; the respirations were deep and rapid; death occurred in coma. No chemical tests were made of the urine. Examination of the body after death did not reveal any striking lesions of any kind; there were small submucous hemorrhages in the stomach and upper part of the small intestine. Microscopically the epithelium of the convoluted tubules showed various stages of degeneration, and in the liver the cells, especially at the periphery of the lobules, appeared to be separated by fluid. Dinitrophenol was demonstrated chemically in the kidney and liver.

The summary of the report of the second case, which occurred in a man, 37 years old, is as follows: Death occurred eleven hours following the oral administration of between eight and seven times the usual therapeutic dose of dinitrophenol. The dose taken was estimated by various methods to have been at least 2.4 but more probably 5 Gm. The man weighed approximately 80 Kg., giving an estimated dosage of 62.5 mg. of dinitrophenol per kilogram of body weight. The drug was self-administered in the apparent attempt to produce hyperpyrexia as a therapeutic measure for a supposed syphilis of the central nervous system. The rectal temperature shortly after death was so high that it could not be recorded by a clinical thermometer, probably being at least 115 F. The onset of rigor mortis was rapid, the body being rigid within ten minutes. A subicteric discoloration of the sclerae and conjunctivae was present and was due in all probability to the color of the drug itself. The anatomic changes consisted chiefly in a marked rigor mortis; an acute pulmonary congestion and edema; ecchymotic hemorrhages in the endocardium, pericardium and pia; mild nephrotic changes in the kidneys, and a slight detachment of the hepatic cells from one another. The clinical and anatomic changes bore a striking resemblance to those seen in cases of heat stroke.

CYTOLGY OF BREAST SECRETIONS. M. KERUBACH AND C. COTUTZ, Deutsche Ztschr. f. d. ges. gerichtl. Med. **22**:235, 1933.

The cytology of the secretions of the breast after miscarriage or labor may be of aid in medicolegal diagnosis. The cell formula of these secretions renders possible the diagnosis of abortion or labor, regardless of retrogressive changes in the uterus. This diagnosis is possible up to three weeks after abortion and longer after labor, and is based on the finding of marked epithelial desquamation. Non-pregnant women show no epithelial elements in the secretions of the breast. There

is a preponderance of epithelial cells in the breast secretions of nonnursing women after pregnancy and after weaning. Tables and illustrations of the constituents of different types of mammary secretions are given.

JACOB KLEIN.

FAT EMBOLISM AFTER TRAUMA AND AFTER BURNING. GEORG STRASSMANN, Deutsche Ztschr. f. d. ges. gerichtl. Med. **22**:272, 1933.

Among 125 persons who succumbed to fatal trauma from blunt forces 8 died suddenly without evidence of fat embolism. However in 17 immediately fatal and 19 rapidly fatal cases fat embolism was demonstrable. When fat embolism was the cause of death there were associated fractures of the long bones, ribs or pelvis. Fat emboli were found in the lungs up to thirteen days after trauma. After burns fat embolism was rare and was mostly associated with bone trauma.

JACOB KLEIN.

MORPHOLOGIC CHANGES IN VISCERA DUE TO DIRECT CURRENT. JULIUS INCZE, Deutsche Ztschr. f. d. ges. gerichtl. Med. **22**:309, 1933.

The direct current causes specific morphologic changes in the parenchyma and stroma of the viscera. For example, the epithelial cells of the renal tubules may assume a characteristic elongated form.

JACOB KLEIN.

SPECTROSCOPIC DEMONSTRATION OF CARBON MONOXIDE IN BLOOD WITH THE AID OF SODIUM STANNITE. O. SCHMITT, Deutsche Ztschr. f. d. ges. gerichtl. Med. **22**:379, 1933.

To 5 cc. of blood 5 small drops of a freshly prepared sodium stannite solution were added, and the blood was examined under the spectroscope. By this means it was possible, as illustrated in the spectrographs, to demonstrate as little as 5 per cent carbon monoxide in the blood.

JACOB KLEIN.

SUCCINIC ACID IN CADAVERS. A. VERDINO, Deutsche Ztschr. f. d. ges. gerichtl. Med. **22**:384, 1933.

From two cadavers examined for organic poisons succinic acid was isolated in considerable amounts. From the one 60 mg. was isolated, from 807 Gm. of tissue; in the other 190 mg. was found in 1,886 Gm. of tissue. The presence of succinic acid is ascribed to bacterial putrefaction.

JACOB KLEIN.

Iso-AGGLUTININ IN OLD BLOOD STAINS. R. WITTE, Deutsche Ztschr. f. d. ges. gerichtl. Med. **22**:397, 1933.

Twenty-three blood stains ranging in age from five to fifty-five years were found to contain iso-agglutinins.

JACOB KLEIN.

SPECTROGRAPHIC EXAMINATION IN ELECTRICAL INJURIES AND GUNSHOT WOUNDS. W. GERLACH, Deutsche Ztschr. f. d. ges. gerichtl. Med. **22**:432 and 438, 1934.

This method of examination is of forensic significance because metallization of tissues and clothing in electrical accidents as well as the presence of various metallic substances in gunshot wounds can be demonstrated readily.

FORMALDEHYDE POISONING. K. BÖHNER, Deutsche Ztschr. f. d. ges. gerichtl. Med. **23**:7, 1934.

A 40 per cent watery solution of formaldehyde causes severe poisoning even in small quantities. In the air passages inflammatory changes develop, with hemorrhage, cell infiltration and edema; in the digestive canal, fixation and hardening of the tissues, also necrotic gastritis and enteritis; in the kidneys, parenchymatous

degeneration. The chief symptoms are dyspnea and cyanosis, dysphagia and vomiting, dizziness and unconsciousness. Death appears to result from respiratory paralysis.

## FROM THE AUTHOR'S SUMMARY.

EFFECT OF COLD AND LIGHT ON THE DETERMINABILITY OF THE BLOOD GROUP.  
B. MUELLER, Deutsche Ztschr. f. d. ges. gerichtl. Med. 23:40, 1934.

Experiments with blood of group A showed that freezing does not materially reduce the determinability of the group. This was the case whether the blood was exposed to cold in the fluid, partially dried or dried state. Sunlight or light from a quartz lamp may reduce the group characteristics materially.

## Technical

COMPARISON OF ASCHHEIM-ZONDEK AND FRIEDMAN TESTS IN NORMAL AND ABNORMAL PREGNANCY. H. C. MACK AND G. H. AGNEW, Am. J. Obst. & Gynec. 27:232, 1934.

Mack and Agnew compared the results of the Aschheim-Zondek test in 546 cases with those of the Friedman modification in 566 cases. The results were almost identical for practical purposes, but the simplicity of technic, the ease of interpretation and the factor of speed strongly favor the method of Friedman. Four cases of choriocarcinoma showed the well known abundance of eliminated hormone. The authors emphasize the need for correlation of the test with the clinical picture, as under circumstances, a negative test may be compatible with an interrupted pregnancy with the products of conception still present in the uterus or tube, and on the other hand, a positive test may be present soon after a termination of pregnancy or in certain abnormal conditions (choriocarcinoma, for example).

I. DAVIDSOHN.

A NEW METHOD OF READING THE FRIEDMAN MODIFICATION OF THE ASCHHEIM-ZONDEK TEST. M. DAVIS, W. KONIKOV AND ELISABETH M. WALKER, Am. J. Obst. & Gynec. 27:274, 1934.

The method is based on the observation of Bercovitz that following the instillation of blood serum of pregnant women into their own conjunctival sacs there follows a pupillary reaction which is absent in nonpregnant women. Davis and his associates observed contraction or dilatation of the pupils in rabbits immediately following the intravenous injection of the urine of pregnancy. The results were compared with the usual changes in the ovaries. There was agreement in 90.6 per cent of the cases of pregnancy, and in 81.8 per cent of the cases of nonpregnancy.

I. DAVIDSOHN.

CONGENITAL SYPHILIS FROM A TRANSFUSION OF BLOOD TO MOTHER DURING PREGNANCY. G. R. WILLIAMSON AND R. A. STRONG, Am. J. Syph. & Neurol. 7: 484, 1933.

A married nurse, four months pregnant, presented cellulitis of her right arm and septicemia, both of which developed in the course of her work, and which necessitated a transfusion of blood. Two transfusions were given, though no Wassermann test was made of the donor. Six weeks later secondary syphilis developed in the patient. There was no reason to suspect syphilitic infection prior to the transfusion. The husband and first child were perfectly normal. This pregnancy resulted in the somewhat premature birth of a 5 pound (2,268 Gm.) congenitally syphilitic infant. The literature is reviewed. The apparent inadequacy of precautions taken in many such cases indicates the need for greater vigilance. At present there is added risk because a less desirable class of professional donors are offering themselves, owing to unemployment in other fields.

JACOB KLEIN.

A SIMPLE INSTRUMENT FOR USE ON THE VERTEBRAL COLUMN. F. HENSCHEN,  
Centralbl. f. allg. Path. u. Anat. **60**:55, 1934.

Henschen describes a powerful, slightly curved chisel, 25 cm. long, with a cutting edge of 8 cm., an eight-cornered handle and a large flat head. The instrument is made of one piece of steel and is driven into the vertebrae with a wooden or hard rubber mallet. The advantages claimed for this device are: its simplicity and ease of manipulation; the clean cuts free from sawdust, and the possibility of removing the spinal cord from the front. GEORGE RUKSTINAT.

A SIMPLE AND RAPID CHEMICAL HORMONAL PREGNANCY REACTION. E. CUBONI,  
Klin. Wchnschr. **13**:302, 1934.

The urine of the pregnant mare contains so little of the hormone of the anterior lobe of the pituitary gland that it cannot be used for the diagnosis of pregnancy by means of the well known reaction in infantile female mice. On the other hand, the urine of the mare contains abundant quantities of follicular hormone. The diagnostic test for pregnancy is based on the injection of the urine into infantile rats or mice, or into castrated mice, and on the observation of a characteristic desquamation of vaginal epithelium. Köber reported (*Biochem. Ztschr.* **239**:209, 1931) the appearance of fluorescence when heated follicular hormone was treated with concentrated sulphuric acid. Cuboni applied that to the diagnosis of pregnancy in the mare. The urine is filtered through filter paper; to 5 cc. of it, 1 cc. of concentrated hydrochloric acid is added and the mixture is heated for ten minutes on a boiling water bath, then it is cooled under running water and 6 cc. of benzene is added. After vigorous shaking (fifty times), the urine is discarded and the supernatant benzene is permitted to settle. Then 3 cc. of the benzene extract is dried by heating at 60-80 C. The sediment is dissolved completely in 0.8 cc. of concentrated sulphuric acid. The solution is heated for a few minutes at 70-80 C. If the test is positive a green fluorescence is shown when the solution is viewed in reflected light. In transmitted light the appearance is (dark red, brown-red or brown) in pregnancy and in nonpregnancy. The performance of the test takes from fifteen to twenty minutes. Another procedure consists in adding 1 cc. of concentrated sulphuric acid to the benzene extract. The mixture is heated for a few minutes on the water bath at 70-90 C., shaken and left for five minutes. Under the colorless layer of benzene there is a layer of sulphuric acid which shows fluorescence in cases of pregnancy. The reaction was correct in all thirty-five cases of pregnancy of mares and in numerous negative controls. The addition of preservative (phenol) did not interfere with the test. Non-preserved urine was satisfactory for the test for more than a month when kept in the icebox, and for fourteen days at 37 C. Specimens which were inoculated with *Bacillus proteus* could also be used. A similar technic permits the carrying out of the test with blood serum.

I. DAVIDSOHN.

THE RELIABILITY OF THE FRIEDMAN RAPID TEST FOR PREGNANCY. F. HEIN,  
München. med. Wchnschr. **80**:1687, 1933.

Hein found the Friedman test quite as reliable as the original Aschheim-Zondek test with the additional advantages of greater speed and easy interpretation. He found, however, that in seven of sixty-five cases, or in 11 per cent, the result was still negative after twenty-four hours, but was strongly positive when read after forty-eight hours. He advises the use for each test of two animals weighing about 2,000 Gm. One of them can be examined after twenty-four hours and if the result is positive, no further observation is necessary, but if the result is negative, the second animal is to be examined twenty-four hours later, at which time the result is final, whether positive or negative.

I. DAVIDSOHN.

# Society Transactions

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AMERICAN SOCIETY FOR EXPERIMENTAL PATHOLOGY

C. PHILLIP MILLER, *Secretary*

*Twenty-First Annual Meeting, Columbia University, New York,  
March 28-31, 1934*

CARL V. WELLER, *President*

THE METEOROLOGICALLY CONDITIONED BIOLOGIC RHYTHM AND EXPERIMENTAL PRODUCTION OF ENDOCARDITIS. WILLIAM F. PETERSEN and A. J. NEDZEL (by invitation), University of Illinois.

When patients are followed day by day clinically or chemically, a definite phase difference in every process can be observed. A phase of decreased activity (A R S) is followed by one of increased activity (C O D). During the former the organism is more alkaline, anabolism preponderates, and with this, reduction is enhanced and spasm is more pronounced in the smooth musculature (high blood pressure; etc.). This phase leads to a definite anoxemia, which in turn initiates the C O D phase, when catabolism, oxidation, increased metabolic rate, relative acidity and dilatation of the blood vessels are all more pronounced. The normal organism pendulates between two such poles. The unstable person reveals evidence of greater amplitude in these rhythmic waves.

The blood pressure curve illustrates the rhythm very well and affords a simple method of observation; periods of pressor increase are followed by distinct lowering of the diastolic blood pressure, not infrequently cumulative and sustained (C O D phase).

During the pressor phase certain tissues of the body become relatively anoxicemic, and this in turn causes stimulation, with a distinct change in the functional status. Regions of potential vascular insufficiency (terminal vessels, epithelial tissues, tissues with sclerotic impairment) are of necessity more often affected than others.

The rhythm is conditioned by many factors—endocrine, emotional, infectious—but perhaps the most important environmental alteration in this region of the world is the meteorological. As a result the changing tide of the A R S and C O D phases is induced largely by meteorological alterations (that is, the cyclonic circulation of the atmosphere). Pressor events are usually associated with a cold or polar front (they may occur, however, with high humidity and heat), and as a result with clinical episodes in the patient—that is, the precipitation of disease is often intimately related to the pressor episodes and the passage of polar fronts.

Inasmuch as bacterial endocarditis is so distinctly seasonal in its onset we have sought to determine whether bacterial localization on the heart valves might follow artificially induced pressor episodes (pitressin). In a series of dogs, streptococci and staphylococci intravenously injected were found to localize on the valves when the animals had been previously given injections of pitressin. Lesions so induced were typical of the ulcerative endocarditis and, in some instances, of the vegetative type of endocarditis that are seen in man (demonstration). The increased demand caused by increased pressure is apparently associated with increased permeability and stickiness of those portions of the valve that come into contact more forcibly, and in such regions bacteria are more liable to localize.

Not only will pressor events, therefore, have the tendency to permit the passage of the bacteria present in the mucous membranes through these mem-

branes, but the same meteorological event or a succeeding one will enhance the possibility of localization of such bacteria on a valve—or in other regions of the body that have been subjected to anoxemic stimulation (spinal cord, giving rise to poliomyelitis; joints, inducing arthritis; dental roots, gallbladder, etc.).

#### HEMORRHAGIC REACTIONS IN NORMAL AND TUBERCULOUS ANIMALS. JULES FREUND, Cornell University.

When certain toxic bacterial products are injected into the blood stream of tuberculous guinea-pigs, the skin at the site of a positive tuberculin reaction becomes hemorrhagic.

In normal guinea-pigs, tuberculin does not prepare the skin to a hemorrhagic reaction.

Tuberculin does not produce an intense or necrotic inflammation in the skin of tuberculous rabbits and fails to prepare the skin for the hemorrhagic reaction.

Nontuberculous guinea-pigs sensitized to horse serum react to horse serum injected into the skin, with redness and edema. Such a reaction is not influenced by a subsequent injection of a potent bacterial filtrate. Tuberculous guinea-pigs sensitized to horse serum react to undiluted horse serum with redness and edema followed by necrosis; the necrosis is sometimes preceded by purple discoloration, an observation described by Dienes. When tuberculous guinea-pigs sensitized to horse serum are given an intradermal injection of diluted horse serum, their reaction becomes hemorrhagic after subsequent injections of bacterial filtrates into the circulation.

As a preparatory factor, diphtheria toxin acts regularly in the normal guinea-pig; silver nitrate, in some of the nontuberculous and in a large proportion of the tuberculous guinea-pigs. Turpentine, broth and aleuronat are inactive in both tuberculous and nontuberculous guinea-pigs.

As factors producing injury, filtrates from meningococci, typhoid bacilli and colon bacilli are active in sublethal doses. Tuberculin is active in tuberculous guinea-pigs when it is used in lethal doses. Witte's peptone, agar and starch produce anaphylactoid symptoms but are as a rule inactive as injury-producing agents.

In the guinea-pig, the skin-preparatory agents have a tendency to produce hemorrhages even without the subsequent injection of the injury-producing agent, and this property may be essential in their action. The production of symptoms of anaphylactoid shocks is not always associated with the production of hemorrhage, as shown by experiments with Witte's peptone, agar and starch.

#### TUBERCULOSIS IN SYPHILITIC AND NONSYPHILITIC RABBITS. JOSEPH D. ARONSON and DAVID R. MERANZE (by invitation), The Henry Phipps Institute, and Mount Sinai Hospital, Philadelphia.

Tuberculous lesions induced by the injection of 0.1 mg. of a culture of the bovine type of tubercle bacillus into the skin of syphilitic and nonsyphilitic rabbits have been studied. Tubercle formation was accelerated and progressed more rapidly in the syphilitic rabbit. Twelve hours after inoculation polymorphonuclears occurred in the tissue about the site of injection as definite discrete aggregations whereas in the nonsyphilitic rabbit polymorphonuclear infiltration was diffuse throughout the subcutaneous tissue. From twenty-four to forty-eight hours after inoculation discrete aggregations of large mononuclear cells with pale-staining nuclei were noted about the site of injection in the syphilitic rabbit. These cells tended to increase, as did the fibroblasts. In the nonsyphilitic rabbit a large, single, circumscribed lesion consisting of polymorphonuclear cells was noted within the first week following infection. In both groups of animals surface ulceration occurred about the second or third week. In the syphilitic rabbit granulation tissue was noted beneath the ulceration, and epithelioid cells were aggregated in groups surrounded by connective tissue. In the nonsyphilitic rabbit the granulation tissue was minimal, caseation was marked, and extensive epithelioid proliferation was present.

tion occurred beneath the ulcer. Caseation was less marked in the syphilitic rabbit, granulation tissue was more marked, and epithelioid cells occurred as discrete aggregations, while in the nonsyphilitic rabbit the epithelioid cells occurred in extensive diffuse areas.

#### OCCURRENCE OF LESIONS IN RABBITS FOLLOWING INJECTION WITH BCG. WILLIAM H. FELDMAN (introduced by F. C. MANN), The Mayo Clinic.

A strain of BCG obtained from Calmette in 1930 and subsequently grown for twenty generations on an egg-glycerin medium was transferred to glycerin-peptone-broth medium and the cultures used to inoculate a series of six rabbits intravenously and four guinea-pigs subcutaneously. One of the rabbits died ten days after inoculation and the other five were killed after one hundred and seventy-four days. Numerous and striking tubercle-like lesions occurred in the lungs of each of the five rabbits. Attempts to cultivate acid-fast bacteria from the lesions were futile, although bacteria of this character were readily demonstrable in appropriately stained sections of the lesions. Emulsions of tissue showing the lesions taken from each of the five rabbits failed to produce demonstrable lesions in other rabbits or guinea-pigs, and attempts to repeat the results in additional experiments have failed.

#### THE FATE OF BCG AND THE ASSOCIATED HISTOLOGIC CHANGES IN THE ORGANS OF RABBITS. MAX B. LURIE (introduced by ESMOND R. LONG), The Henry Phipps Institute, Philadelphia.

Three series of rabbits were inoculated intravenously with 1 mg. of a culture of a strain of BCG obtained from the Pasteur Institute. At various intervals organs, blood and bile were cultured on Löwenstein's egg medium modified by the addition of bone marrow extract. Tissues adjoining those cultured were studied histologically.

The greater the primary deposition of bacilli the more rapid was the initial growth of the bacilli and the earlier the beginning of their destruction. With the smallest original deposition, however, destruction began immediately in the lung and the liver, but at the same time multiplication occurred in the spleen and the lymph nodes, indicating local immunity of the lung and the liver.

The growth of the bacilli is associated with a local multiplication of mononuclears by mitosis, their accumulation into nodules, the destruction of the bacilli and a formation of epithelioid and giant cell tubercles. Necrosis of invading polymorphonuclears, exudation of fluid and cells into the alveoli and caseation occur with the development of hypersensitivity to tuberculin. By the fourth week, when this was marked, bacilli had been almost completely destroyed in all organs and lymph nodes.

All the tuberculous changes were usually resolved completely by the second month; yet isolated bacilli persisted in lymph nodes even fourteen months after inoculation.

#### EFFECT OF FERRIC CHLORIDE IN EXPERIMENTAL TUBERCULOSIS. VALY MENKIN, Harvard Medical School.

Previous studies had shown that repeated intravenous injections of 0.25 per cent ferric chloride in rabbits infected with bovine tubercle bacilli were followed by an accumulation of iron in the tubercles (chiefly within the caseous areas). Concomitantly there was a retardation in the development of the disease, as evidenced both by an increase in the survival time and by a less extensive tuberculous involvement in the treated as compared with the nontreated animals. These results were obtained in two independent series of experiments.

These studies have been continued. In the present series virulent bovine tubercle bacilli (Ravanel) were inoculated subcutaneously instead of intravenously as in the previous experiments. Treatment with ferric chloride was started *promptly* after

inoculation of the bacilli and was continued for about four months. Five controls lived from 91 to 170 days, with an average survival time of 130 days. Four experimental rabbits survived from 112 to 326 days. One experimental rabbit was killed 344 days after inoculation. The average longevity of the experimental rabbits was 246 days, an increase of 89 per cent over the survival time of the controls.

Exactly the same type of experiment was repeated in ten rabbits which, however, had been previously inoculated by intravenous injection of a relatively avirulent strain of bovine tubercle bacilli (Cernay), in order to see whether partial immunity induced by the previous infection would enhance the retarding effect of ferric chloride. Such data would be of importance in reference to adult human tuberculosis, which is doubtless the tuberculosis of reinfection. The five controls lived between 47 and 130 days, with an average survival time of 81 days. Three of the experimental rabbits died 81, 110 and 131 days after subcutaneous reinoculation with virulent tubercle bacilli. Two of these animals succumbed to infections of the upper respiratory tract. The two remaining experimental rabbits progressively increased in weight and lived for over six months after the death of the last control animal. On the three hundred and thirty-fourth day after reinoculation they were killed. The extent of the tuberculous involvement in these two animals was far below that found in the last survivors of the control group. The average longevity of the experimental rabbits was 198 days, an increase of 144 per cent over the average survival time of the controls. By employing the subcutaneous route of infection and instituting intravenous ferric chloride treatment immediately, more pronounced effects were obtained than in the previous series. In vaccinated animals the results seemed to be even more striking than in the non-vaccinated group.

The mechanism of the favorable effect of ferric chloride is now under investigation. The frequent finding in the lungs of treated animals of zones of engorged capillaries about tubercles, at times associated with some fibroblastic proliferation, suggests the end-stage of a superimposed inflammatory reaction induced by the accumulation of iron in the tubercles. Ferric chloride is in itself an intense inflammatory irritant. The lungs of animals surviving long after the cessation of treatment were often characterized by moderate connective tissue proliferation at the peripheries of tubercles. These fibrous zones, in their location with respect to the tubercles, corresponded in a general way to the areas of engorgement found at an earlier stage. Similar areas of congestion were observed in a comparatively small percentage of the control tuberculous rabbits. The hypothesis is being subjected to further study.

It is noteworthy that in experimental rabbits, hemosiderin, or at any rate a substance indistinguishable from it by the usual tests, is found in great abundance in granular form within the mononuclear phagocytes of the spleen, in the Kupffer cells of the liver, in the reticular cells of the bone marrow, within the tubules of the kidney and sometimes in the cells of the alveolar walls in the lungs. These animals had no anemia. This raises the question whether hemosiderin is necessarily a product of the partial degradation of hemoglobin, or whether the mere cellular release of iron, which in turn may combine with some of the plasma proteins, is not sufficient to account for the formation of hemosiderin. Furthermore, it is conceivable that the hemosiderosis observed in experimental animals may also play a part in the favorable effects of ferric chloride on the course of tuberculosis in rabbits by activating and causing a new formation of cells of the reticuloendothelial system. That this may be suggested by the following observations: Variable degrees of fibrosis at the peripheries of malpighian follicles were found associated with extensive hemosiderosis in the spleen. In the pulmonary lesions of the experimental rabbits many mononuclear cells at the peripheries of casedated centers were occasionally seen to be loaded with iron-staining material. Two normal rabbits that had received repeated intravenous injections of 0.25 per cent ferric chloride for one month were subsequently inoculated intravenously with

virulent bovine tubercle bacilli. They survived 115 and 124 days respectively, while seven tuberculous rabbits that had received no such preliminary treatment survived on the average 54 days, the longest survivor dying on the sixty-seventh day of its disease. This subject is now being investigated further.

**EXPERIMENTAL PULMONARY EMBOLISM AND INFARCTION.** BERNHARD STEINBERG and CARL S. MUNDY (by invitation), Toledo Hospital, Toledo, Ohio.

Pulmonary emboli were produced in dogs by introduction of lead shot into their jugular veins. At varying intervals the emboli were followed by injections of iodized poppy-seed oil 40 per cent. Complete closure of the lumen of the vessel was produced. Hemorrhagic infarcts appeared involving parts or the whole of a lobe. These infarcts at the sixth or seventh day began gradually to disappear with a return of the tissue of the lung to an approximately normal state except for moderate interstitial fibrosis. At varying intervals following the establishment of pulmonary embolism, iodized oil was injected into the bronchial circulation. During the period in which the infarcts were most prominent, the bronchial circulation in the infarcted areas was not very apparent. On the fifth day, the bronchial circulation became significant, and coincidentally with the return of the infarcted lung to a comparatively normal state the bronchial circulation was very prominent, resembling in extent the pulmonary circulation. Although the present experiments do not establish either the independent blood supply or the free anastomosis between the two circulations, indirect evidence points to the bronchial arteries as independent in the assumption of the circulation in the infarcted area.

Although as many as three hundred and fifty shot were introduced into the pulmonary circulation and the dogs were kept for a year and a half, at no time was there any untoward symptom or death due to embolism.

**THE MOVEMENT OF PARTICULATE MATTER ON THE MUCOUS SURFACES OF THE TRACHEA AND BRONCHI.** R. Z. SCHULZ (introduced by S. B. Wolbach), Harvard Medical School.

The transportation of minute particles of carbon and garnierite in the trachea and bronchi has been observed in guinea-pigs, rabbits, cats, dogs and chickens. The general movement is from the bronchi toward the larynx with a slow and, at times, imperceptible movement in the bronchi and a more rapid propulsion in the trachea. The rate at which particulate matter is carried along the mucous surfaces varies with the different species tested, being slowest in guinea-pigs and chickens and increasingly more rapid in rabbits, cats and dogs. The factors which appear to influence the rate of movement are the amount and the tenacity of the film of mucus or fluid and the activity of the cilia. In animals infected with *Streptococcus haemolyticus* or *Bacillus bronchisepticus*, in which the exudate is of a seropurulent type, the rate of movement is greatly increased. Alcohol, ether and phenobarbital sodium, in narcotizing doses, do not appear to alter the activity of the cilia. Chloroform causes a slowing and early cessation of activity.

These studies indicate that one of the important functions of the ciliated cells of the trachea and bronchi of the mammal is to facilitate the mucous glands in maintaining an even film of mucus on the lining epithelium. A second function is to aid in the elimination of particulate material from the tracheobronchial tree.

**AN ATYPICAL REACTION TO VACCINE VIRUS IN THE RABBIT.** LOUISE PEARCE, C. K. HU (by invitation) and PAUL D. ROSAHLN (by invitation), The Rockefeller Institute for Medical Research, New York.

The reappearance of rabbit pox last December approximately ten months after the subsidence of a severe epidemic of the disease necessitated the immediate application of methods designed to protect a rabbit-breeding colony of some one thousand and eight hundred animals. A considerable number of the older rabbits had recovered from the disease and might still be immune, but there were many

young adults and a large number of young born after the epidemic which presumptively were susceptible. On the basis of the results of experiments in which it was found that immunity to vaccine virus afforded some protection against rabbit pox, the colony was vaccinated with New York City Board of Health virus which had been propagated in tissue culture by Dr. T. M. Rivers. The method of vaccination employed was the intradermal injection of virus at a single site.

This mass vaccination resulted in several types of reactions which, according to generally prevalent opinion, are atypical. They occurred so frequently, however, as to raise the question whether current conceptions of vaccinal reactions are sufficiently inclusive.

In the susceptible adults, the expected local reaction developed, and in a number of cases a few scattered papules in the shaved skin surrounding the site of vaccination were observed. But, in addition, a generalized papular eruption occurred not infrequently, the lesions being distributed in portions of the body remote from the local reaction, for example, the ears. Other symptoms, such as generalized adenitis and orchitis in male animals, were common. Many of these rabbits were obviously ill, and some of them critically so, but there were only three fatalities.

In the young stock, all presumably susceptible, the results were sharply divided into two classes, one comprising typical local reactions to vaccination, and the other, results in which no local reaction was seen. In the first group there were a number of reactions associated with generalized eruptions corresponding to that which obtained among the adults, but the condition was in general more severe and the mortality considerably higher. In the rabbits in which no local vaccinal reaction was observed generalized lesions were comparatively frequent and severe and the death rate was high. Furthermore, there were several fatal cases in which no clinical evidence of vaccinal infection had been observed, either local or general, but in which postmortem examination revealed a more or less widespread distribution of vaccinal lesions in the viscera. Finally, there remained a group of young, presumably susceptible rabbits which gave no clinical indication, either local or general, of any reaction to the vaccination.

There are many aspects of these results which should be briefly referred to since the situation is by no means a simple one. The character of the reaction in nursing young was apparently influenced by the immune or nonimmune state of the doe, and the reaction of different breeds was not identical. There was likewise a sex difference, the reaction being generally milder in females than in males and especially so in pregnant and nursing does. The relation of the type of reaction to the size of the dose of vaccine virus administered must also be considered. These and other points as well must be taken into account before a final appraisal of the results can be made.

It is evident, however, that the results of our experience do not support the opinion that in the rabbit a wide distribution of lesions after local vaccination is unusual. Generalized vaccinia may be frequent, at least in certain circumstances, and, furthermore, the clinical manifestations of the condition are sufficiently distinctive to differentiate it as a disease entity.

#### SPORADIC ENCEPHALITIS IN COWS. F. S. JONES AND RALPH B. LITTLE (by invitation), The Rockefeller Institute for Medical Research, Princeton, N. J.

Since 1930 a disease clinically characterized by well defined nervous symptoms has been studied. The brains of thirteen affected cows have been obtained. In all instances except one the animals were slaughtered between the second and sixth days of the attack. The incidence in the herd has been low, but a single case is apt to occur in a given group over a period of years. Adults of both sexes are affected.

Gross lesions either in the viscera or in the central nervous system have not been observed. Examination of fixed and stained material reveals well defined changes localized principally in the midbrain, stem and anterior portion of the cord. These consist in tiny areas of softening. Early in the disease the lesions consist of a

loosely arranged accumulation of polymorphonuclear leukocytes; later the polymorphonuclears become more numerous, and round cells appear. When the disease is well advanced the lesions are principally composed of round cells. Well defined perivascular infiltrations are encountered in the vicinity. These infiltrations, like the areas of softening, change in character and finally become almost purely round-celled. Cortical and meningeal lesions are infrequent.

Although the disease resembles others caused by filtrable agents, thus far it has not been possible to demonstrate such an agent. The inoculation of suspensions of affected midbrain and stem intracerebrally into calves, rabbits or guinea-pigs results in an acutely fatal meningitis. From both the original material and the brains of the experimental animals a gram-positive rod is readily cultivated. A similar organism is readily identified in the earliest lesions of the disease (microscopic areas of softening) in the fixed and stained preparations.

Intracerebral inoculation of calves, guinea-pigs and rabbits with a pure culture of the organism as a rule results in fatal meningitis. When extremely small doses are given, or the animal's resistance is stimulated by a previous intranasal inoculation, encephalitis may result.

THE PATHOGENICITY OF SWINE INFLUENZA VIRUS FOR FERRETS. RICHARD E. SHOPE (introduced by Carl Ten Broeck), The Rockefeller Institute for Medical Research, Princeton, N. J.

The observation of Smith, Andrewes and Laidlaw that the virus of swine influenza is pathogenic for ferrets when administered intranasally has been confirmed. A disease that is clinically more severe and pathologically more extensive than that described by the aforementioned workers is obtained if the ferrets are anesthetized with ether prior to infection. Animals infected in this way show at autopsy an edematous type of pneumonia of lobar distribution which may sometimes terminate fatally. The virus is easily transmitted serially through ferrets, and it maintains its pathogenicity for this species when stored in 50 per cent glycerol at refrigerator temperature for as long as seventy-five days. After serial passage through twelve ferrets, the virus is still capable of inducing swine influenza when mixed with *Haemophilus influenzae-suis* and administered intranasally to swine. Passage through ferrets causes no apparent attenuation of the virus for swine. Serum from pigs recovered from swine influenza is capable of neutralizing the ferret-passed virus for either swine or ferrets. Likewise, serum from recovered ferrets neutralizes the swine influenza virus for either ferrets or swine.

MATERNAL TRANSMISSION OF VACCINIAL IMMUNITY IN SWINE: II. THE DURATION OF ACTIVE IMMUNITY IN THE SOW AND OF PASSIVE IMMUNITY IN THE YOUNG. JOHN. B. NELSON (introduced by Carl Ten Broeck), The Rockefeller Institute for Medical Research, Princeton, N. J.

Two sows which had previously been vaccinated with vaccinia virus have continued to transmit immunity to the suckling young of six successive pregnancies over a period of three years. Pigs from the sixth litter were as well protected as those farrowed at the end of the first pregnancy after vaccination.

Vaccination of the suckling young at successive age intervals showed that the maternally acquired immunity began to decline during the second month of life and in most cases had practically disappeared by the end of the third month. The virus used in testing the suckling pigs, during the first week after birth, exerted little or no immunizing effect.

THE KUPFFER CELL IN RELATION TO IMMUNITY TO THE VIRUSES. J. W. BEARD (by invitation) and PEYTON ROUS, the Rockefeller Institute for Medical Research, New York.

Kupffer cells containing particles of iron have been collected with the magnet and tested for their influence on the virus of vaccinia. The living cells neutralized

the virus, whereas killed ones were without effect. Similar findings were obtained with the clastmatocytes of aleuronat exudates, not only with the virus of vaccinia but with that causing the Shope fibroma. Polymorphonuclear leukocytes, on the other hand, whether alive or dead, were without any neutralizing effect on vaccinia, and living cells of this sort seemed to enhance the activity of the Shope virus.

THE HIGH PATHOGENICITY OF A RECENTLY ISOLATED STRAIN OF SPIROCHAETA PALLIDA. C. K. HU (introduced by L. Pearce), The Rockefeller Institute for Medical Research, New York.

A strain of *S. pallida* isolated in 1931 from an inguinal lymph node of a Chinese patient suffering from a profuse maculopapular syphilitic eruption was found to be highly pathogenic in its early passages in the rabbit. The clinical manifestations of disease induced by this strain in the first three passages were found to be of the same order as those of older strains which had been propagated in the rabbit for hundreds of generations.

Two experiments were carried out in March and October 1932, at which time the strain was in the fourth and sixth generations following passage through animals. In each experiment ten male rabbits were inoculated intratesticularly and ten in the skin of the prepuce. The results of the two experiments were practically identical. Furthermore, comparable results were obtained by the two different routes of inoculation.

The pathogenicity of the new strain was measured by the incidence, the time of occurrence and the character of the primary lesions, together with the incidence, the time of occurrence, the distribution and the character of generalized lesions including metastatic orchitis and lesions of the skin, periosteum and bones, and eyes. The results obtained in thirty-six of the forty animals that survived the observation period of three and a half months may be summarized as follows: The incidence of primary lesions was 100 per cent, with an average incubation period of 18.3 days; metastatic orchitis in the uninoculated testicle occurred in 88.9 per cent of the animals in an average period of 52.2 days after inoculation; the average relative incidence of generalized lesions was 7.4 lesions per animal; the average period of activity of the lesions, as measured by the difference between the mean time of the first and that of the last lesion, was 23.5 days.

These results were entirely comparable with those obtained in similar experiments carried out at the same time and under the same experimental conditions. In these experiments, four strains of *S. pallida* were used, namely, the Nichols, the Zinsser-Hopkins, and two older Chinese strains, all of which have been carried in rabbits for many years.

It would appear, therefore, that the capacity of a strain of *S. pallida* to induce in the rabbit the characteristic disease picture of experimental syphilis including a diversity of generalized manifestations is not necessarily dependent on a long sojourn of the strain in the new host, in which circumstances it has been presumed to have become "adapted" and thereby to be capable of the production of disease.

ARSENOXIDE (META-AMINO-PARA-HYDROXYPHENYL ARSENOXIDE) IN EXPERIMENTAL ANIMALS. O. M. GRUHNIT, Parke, Davis and Company, Detroit.

The therapeutic and the toxic effects of the arsphenamines are thought to be due to the action of their "breakdown" product, the arsenoxide (meta-amino-para-hydroxyphenyl arsenoxide), known under the specific name of mapharsen. The compound has been recently introduced in the treatment of spirochetosis. Its toxicologic and pathologic effects are presented here in brief.

The tolerated dose of mapharsen is from 10 to 22 mg. per kilogram according to the animal used. In the white rat the intravenous tolerated dose is from 18 to 20 mg. per kilogram. Rabbits tolerate from 10 to 14 mg. per kilogram. In rats, rabbits and dogs, an intravenous dose of from 1 to 2.5 mg. per kilogram causes no gross or microscopically demonstrable lesions in organs. A dosage of 3, 4 and 5 mg. per kilogram a week, or a total of 76 mg. per kilogram in a period of

fifteen weeks, in dogs causes occasional temporary albuminuria, but no organic changes such as result in abnormalities of the total nonprotein nitrogen or sugar content of the blood.

A dosage of from 6 to 8 mg. per kilogram causes albuminuria, mild congestion of the liver and cloudy swelling of the cortical tubular epithelium of the kidneys. The maximal tolerated and the minimal lethal dose (60 per cent respectively of the animals live or die) may induce from mild to severe changes in the kidneys and mild injury in the liver, spleen and other organs. The predominating changes in the organs of rabbits from the maximal tolerated and minimal lethal doses appear in general as follows:

At the end of twenty-four hours following a treatment all organs show moderate degrees of congestion. No focal hemorrhages or necrosis are present in the liver. The liver appears edematous; some of the hepatic cells are granular, and some appear partly lysed. The kidneys are congested and edematous and some tubules of the cortical zone show coagulation necrosis. The glomerulae appear congested.

At the end of seventy-two hours the liver is somewhat congested and edematous, but no focal necrosis or hemorrhage is noted. In the kidneys the coagulation necrosis is distinct and is limited to the cortical zone.

At the end of from five to seven days the pathologic changes have reached their maximal development. The liver appears almost normal. The kidneys show in place of the coagulation necrosis an extensive deposit of calcium in the tubules of the cortical zone. The glomerulae appear congested; the distal collecting tubules may contain casts, but otherwise the collecting tubules appear well preserved. The lungs of animals dying at this stage are congested, and the alveoli are filled with serous exudate. No coagulation thrombi are noted.

As the duration of life of the animal extends beyond the seven day period, the reparation of the tissue begins to take place, namely, decalcification in the kidneys. On the twelfth day the deposits of calcium have receded to include only the intermediate zone of the cortex. At the end of twenty-four days almost all the calcium has disappeared from the tubules, and fibrosis has taken the place of the necrotic tubules. At the end of thirty-six days the kidneys in a number of animals appear almost normal, except for increased fibrosis and a reduced number of tubules. In other animals the necrotic tubules have been replaced by deposits of fat.

In summary, mapharsen causes only slight injury to the liver even with toxic doses. It is strongly nephrotoxic in minimal lethal doses and in general causes coagulation necrosis of the cells of the cortical tubules. The necrosis is followed by deposition of calcium which in turn, on healing, is replaced by fibrosis or fat. The clinical dosage of from 1 to 1.5 mg. per kilogram causes neither histologic changes in the organs nor disturbance in the constituents of the blood.

The histologic changes, in general, resemble those produced by the arsphenamines, except that the liver at no time appears so severely injured as in the case of the latter.

#### FURTHER STUDIES ON PREVENTION OF SERUM SICKNESS. MOYER S. FLEISHER and LLOYD R. JONES (by invitation), St. Louis University.

Treatment of whole serum or of the pseudoglobulin fraction of serum by lipid solvents may markedly diminish its activity in causing serum sickness in rabbits. This treatment does not alter the solubility of the dried protein in physiologic solution of sodium chloride when special consideration is given to the choice of solvents and to the factors of time and temperature.

Studies have been made of the effect of this treatment of serum or serum fraction on its antibody content.

The factor or factors extracted by the lipid solvents have been investigated.

TREATMENT OF TYPHOID FEVER WITH ANTITYPHOID SERUM. GREGORY SHWARTZMAN, GEORGE BAEHR (by invitation) and WILLIAM Y. HOLLINGSWORTH (by invitation), Mount Sinai Hospital, New York, United States Marine Hospital, New Orleans, and Charity Hospital, New Orleans.

The phenomenon of local skin reactivity to bacterial filtrates made possible a demonstration of *Bacillus typhosus* toxic substances which are identical or closely related to true exotoxins and also made it possible to develop antitoxic serum which neutralized these toxic substances specifically and in multiple proportions. Serums of high neutralizing and agglutinating titers prepared by prolonged immunization of horses with *B. typhosus* toxins were used for the treatment of eighty-four patients with typhoid fever.

As controls, seven patients were treated intravenously with from 300 to 500 cc. of normal horse serum. Four died and three recovered. In only one case did the clinical course appear milder after the patient had received serum. Injection of large amounts of normal horse serum seemed to increase the severity of the disease and may have been a factor in the high death rate in the control series. In the series of patients treated with antitoxic serum, from 100 to 500 cc. was injected intravenously in divided doses over periods of from thirty-six to seventy-two hours. In forty of the eighty-four patients there was observed a striking influence on the toxemia, fever, bacteremia and duration of the disease. In thirteen patients there was a definite effect on the toxemia but no striking effect on the duration of the fever. In thirty-one there was no convincing influence of the serum on the course of the disease. Twelve of these received either inadequate amounts of serum, serum preparations of low antitoxic titer early in the work, treatment while moribund, or treatment within thirty-six hours of death from pneumonia. Of the eighty-four treated patients, seven died. Of these, one was moribund when serum was being given; one died of pneumonia within thirty-six hours after receiving the serum; two were given an early serum of low neutralizing potency; one died of pneumonia two weeks after the administration of serum; one died of perforation after a brilliant effect of the serum on the toxemia and fever. Only one patient died of toxemia (within twelve hours after the administration of serum).

THE CHEMOTROPIC ATTRACTION OF HUMAN LEUKOCYTES BY MICRO-ORGANISMS AND VARIOUS SUBSTANCES. MORTON McCUTCHEON and HAROLD M. DIXON (by invitation), The University of Pennsylvania.

The chemotropic response of human polymorphonuclear leukocytes to different types of micro-organisms was evaluated through experiments in vitro. Under the microscope the net distance over which each cell approached a clump of bacteria was measured, and this distance was divided by the total length of the path of the cell. The resulting quotient was a measure of the chemotropic response, having as extreme values +1 if the cell moved directly toward the bacteria, and -1 if the cell moved directly away from them. With staphylococci, streptococci, pneumococci, typhoid and tubercle bacilli, *Micrococcus tetragenus* and certain yeasts, the mean quotients ranged only from +0.73 to +0.86, indicating approximately equal attraction under these conditions. With the yeast *Torula histolytica* the quotient was +0.57. Control leukocytes, wandering in fields free from known chemotropic influence, gave a value of +0.07. With substances other than bacteria a wide range of values was obtained: gelatin, 0.0; dried blood, +0.18; dried leukocytes, +0.25; starch paste, +0.71.

THE PRODUCTION OF ANTIBODIES WITHIN LYMPH NODES. PHILIP D. MCMASTER and STEPHEN S. HUDACK, The Rockefeller Institute for Medical Research, New York.

*The Formation of Agglutinins Within Lymph Nodes.*—When a suspension of killed *Bacillus paratyphosus* B was intradermally injected, on two successive days, in one ear only of a mouse, an extract from the node draining the lymphatic of that

ear a week later showed agglutinin in high concentration. Agglutinin was present, too, in the blood in lower concentration. None was found in the lymph node of the opposite side. The experiment was repeated in a large group of mice.

To evoke equivalent inflammatory reactions on both sides, a killed culture of *B. paratyphosus* B was injected intradermally into the right ear and Schick test toxin into the left. A week later agglutinin for *B. paratyphosus* B was found in the extract from the node on the side receiving the organisms, at a dilution of 1:240. It was present in the serum at a dilution of 1:60. None was found in the extract of the node of the opposite side.

In other experiments the same antigens were used but three hours later the ear inoculated with paratyphoid bacilli was amputated to avoid a possible drainage to the lymph node of antibody formed at the site of injection. Nine days later agglutinin was found in the extract of the lymph node on the side receiving the bacilli at a dilution of 1:120, and not in that of the lymph node on the other side. The serum gave a positive reaction at the dilution 1:60.

Killed cultures of *Bacillus enteritidis* and of *Bacillus prodigiosus* were injected intradermally into the ears of mice, the former on the right side, the latter on the left. From four to sixteen times as much agglutinin for *B. enteritidis* was found in the extracts from the nodes of the ears receiving this bacterial vaccine as in the extracts from the nodes of the other ears, and from three to ten times as much as in the serum. Similarly agglutinin for *B. prodigiosus* was found in far higher concentration in extracts of the nodes of the ears receiving these bacilli. Much less was present in the blood and still less or at most an equal concentration in the nodes on the other side. The lymph nodes on both sides were inflamed to about the same extent.

The longer the interval of time between the injection of the antigen and the examination for antibody up to three weeks, the greater was the concentration of the latter in both nodes and serum. With the passage of time the concentration in the serum equaled that in the nodes but did not exceed it.

*A Comparison of the Formation of Agglutinin in the Lymph Nodes of Resistant and Susceptible Mice.*—Highly resistant and susceptible mice of closely inbred strains, the reactions of which have been extensively studied by Webster and his collaborators, were obtained for the work through the kindness of these investigators.

Killed cultures of various agglutinin-forming bacteria were intradermally injected into the ears of groups of animals from these strains and a third mongrel strain, only one antigen being used for each experiment. After varying intervals of time the cervical lymph nodes draining the lymphatics of the ears were removed and the animals were bled for serum. The lymph nodes were extracted and the agglutinin titers of the extracts and of the serums compared in the various groups.

Almost invariably the concentration of agglutinin was slightly greater in the lymph nodes of the susceptible animals than in those of the resistant ones. So, too, in the case of the serum. The resistant group and the mongrel group formed approximately the same amount of agglutinin.

The mice of the strain manifesting a general lack of resistance seemed capable of forming slightly more specific agglutinin than those of the resistant strain.

#### GASTRIC SECRETION FOLLOWING IRRADIATION OF THE EXPOSED STOMACH AND THE UPPER ABDOMINAL VISCERA BY ROENTGEN RAYS. ALBERT M. SNELL and JESSE L. BOLLMAN, The Mayo Clinic.

Irradiation of the stomach is one of the methods that has been proposed for reducing gastric acidity in the treatment of peptic ulcer in man.

We have studied a series of normal dogs to determine the effect of irradiation on the gastric response to test meals and to stimulation with histamine. Irradiation of the exposed stomach was followed by a marked decrease in gastric acidity, and in some animals complete anacidity was produced temporarily. Irradiation of the upper portion of the abdomen also produced a temporary reduction in gastric acidity, but only occasionally was there a complete disappearance of free

hydrochloric acid. Irradiation of the extremities produced a definite but less extensive reduction in gastric acidity. After extensive irradiation of the stomach, sections of the gastric mucosa revealed no significant pathologic changes. We have been unable to produce permanent anacidity, in spite of the fact that irradiation was carried to the limits of tolerance. The dogs have been observed over a period of a year.

REESTABLISHMENT OF CORONARY CIRCULATION IN PROGRESSIVE CORONARY OCCLUSION. HAROLD F. ROBERTSON (introduced by ELLIOTT C. CUTLER), Peter Bent Brigham Hospital, Boston.

In coronary occlusion, the vitality of the heart muscle may possibly be maintained by a supply of blood from anastomosing coronary vessels, from rami telae adiposae, from vasa vasorum of the aorta, from backflow in thebesian vessels, or from backflow in the coronary sinus; or, when early panthoracopericardial adhesions are present, their vessels may help to supply the cardiac wall. Also, biochemical adjustments may sustain a heart when its normal vascular supply is diminished.

In a series of dogs an experiment was undertaken to determine which vessels might nourish the cardiac wall after the coronary vessels had been gradually occluded by ligature in stages. There being evidence that primary ligature of the cardiac veins gives optimum opportunity for a dilatation of coronary and thebesian vessels, this was first done in stages. A test of the theory that the heart muscle is supplied from a coronary sinus backflow was thus automatically omitted. The arteries were then tied in a series of operations, small vessels being ligated first. Finally, the vascular panthoracopericardial adhesions formed during the course of the experiment were separated, the animals usually dying as a result of fresh infarction of the cardiac wall following the separation of the adhesions.

As in many cases the heart tolerated coronary occlusion as long as these adhesions were preserved, it appears that their progressive development supplied the heart to a great extent. No increase was found in anastomoses of coronary vessels, rami telae adiposae or aortic vasa vasorum; in some cases thebesian vessels were found dilated. The possible ability of the former channels to proliferate or of dilated thebesian vessels to function in compensation for a progressive coronary occlusion was not evident, but may have been taken over by the unavoidable formation of the vascular adhesions.

Slides of hearts in gross are presented to show vascular adhesions, and sections, to show dilated myocardial vessels.

STUDIES OF EXPERIMENTAL CORONARY OCCLUSION. ROBERT TENNANT JR. (introduced by RAYMOND HUSSEY), Yale University.

After ligation of the anterior descending branch of the left coronary artery in dogs the lactic acid content of heart muscle from the affected zone was increased from 100 to 200 per cent over that of control muscle from the right ventricle, irrespective of the duration of the period of ligation up to twenty-four hours. The glycogen content of muscle from similar zones was decreased in every instance. In a second group, in which reestablishment of circulation was effected by removal of the ligatures after intervals varying from a half hour to eight hours, and the animals were sacrificed two hours later, the lactic acid content of muscle from the involved zone remained lower than that of muscle from the control zone, as did also the glycogen content. No histologic changes were observed in the myocardium of any of the animals in the first group up to twelve hours. On the other hand, in the second group, in which the circulation was reestablished, there were extensive hemorrhage, edema, polymorphonuclear leukocytic exudate and slight necrosis throughout the involved zone. In both groups the electrocardiographic changes consisted of T-wave alterations, ectopic premature ventricular contractions, ventricular tachycardia and ventricular fibrillation. The premature contractions appeared within from six to eight hours after the ligation of the vessels; in the second group they appeared earlier.

**THE NATURE OF THE INTIMA AS A TISSUE.** ELIZABETH M. RAMSEY and DAVID W. GAISER (introduced by RAYMOND HUSSEY), Yale University.

A strain of *Streptococcus viridans* known to be pathogenic for rabbits was injected into the rabbit carotid artery between two ligatures. The observations on a series of rabbits were as follows: Six hours after the injection a polymorphonuclear leukocytic exudate was seen in the adventitia of the vessel. At twelve hours the exudate had reached the midportion of the media. At from twelve to twenty-four hours the exudate was present throughout the wall of the vessel and particularly densely accumulated just outside the internal elastic membrane. Between twenty-four and seventy-two hours the exudate was seen also in the lumen of the vessel, and the elastic fibers of the media were separated by dense accumulations of leukocytes. In the later stages, necrosis of fixed tissues became apparent associated with the exudate. At all periods there was cellular infiltration in the connective tissue surrounding the vessel. As the exudate in the wall of the vessel became more widespread and more dense there was also an increase in the amount of exudate in the periarterial tissues, but the infiltration was always most marked in the wall of the vessel. Bacterial stains showed large numbers of organisms in the lumen of the vessel, but none were seen elsewhere. Blood vessels in control animals subjected only to the operative procedure and to double ligation for equal periods of time showed no detectable changes in the wall of the vessel in stained preparations and only slight hemorrhage and minimal cellular infiltration in the periadventitial tissues.

These observations seem to indicate that diffusible substances associated with the streptococcus penetrate the wall of the vessel from within and call forth a leukocytic response. The cells of the exudate apparently issue from the periadventitial vessels and progressively infiltrate the whole wall, finally filling the lumen.

**EARLY INCIDENCE OF SPONTANEOUS MEDIAL DEGENERATION ("ARTERIOSCLEROSIS") IN THE AORTA OF THE RABBIT.** H. D. KESTEN, Columbia University.

This article will appear in full in a later issue of the ARCHIVES.

**FUNCTIONAL HYPERTROPHY OF THE KIDNEY.** J. L. BOLLMAN and F. C. MANN, The Mayo Foundation.

This article will appear in full in a later issue of the ARCHIVES.

**THE TITRATION OF PROTHROMBIN IN CERTAIN PLASMAS.** E. D. WARNER (by invitation), K. M. BRINKHOUS (by invitation) and H. P. SMITH, State University of Iowa.

If one adds to plasma an optimum amount of calcium plus an excess of thromboplastin, the prothrombin present will be converted completely into thrombin. By suitable procedures one can determine the relative amount of thrombin present and hence the relative amount of prothrombin in the original plasma. Certain pitfalls in this titration of prothrombin are discussed. An excess of antithrombin, especially, interferes with the estimation of thrombin. The antithrombin can be eliminated by ammonium sulphate fractionation.

Experiments on dogs show that in from twenty to thirty hours after chloroform poisoning the plasma prothrombin has fallen to less than 5 per cent of normal. Recovery occurs within a week. The fall in plasma fibrin is less extreme. The rise to normal is parallel to the rise in prothrombin.

Peptone plasma, after being freed from its antithrombin, can be shown to have a normal content of prothrombin.

THE UTILIZATION OF PLASMA PROTEIN (DOG AND HORSE) AND HEMOGLOBIN GIVEN BY VEIN. WESLEY T. POMMERENKE (introduced by GEORGE H. WHIPPLE), The University of Rochester, Rochester, N. Y.

Evidence is presented showing that the level of the circulating plasma protein in dogs can be raised to a value about 50 per cent above the normal by intravenous injection of heparinized dog plasma. This seems to produce no untoward results. When sugar is also given by mouth, the animals can be maintained practically in nitrogen equilibrium. The surplus protein is removed from the circulation and does not escape in the urine. This seems to point to a utilization of the administered protein in the bodily economy.

When horse plasma or dog hemoglobin is given intravenously under identical conditions, there is little evidence that these proteins are utilized to spare body protein.

FORMATION OF INTERCELLULAR SUBSTANCE BY ADMINISTRATION OF ASCORBIC ACID (VITAMIN C) IN EXPERIMENTAL SCORBUTUS. S. BURT WOLBACH, VALY MENKIN and MIRIAM F. MENKIN (by invitation), Harvard Medical School.

Scurvy is characterized by a cessation in the normal formation of intercellular substance on the part of supporting tissues. This was established by the earlier studies of Wolbach and Howe (1926). The immediate effect of vitamin C administered in the form of orange juice to guinea-pigs was a prompt reparative response. This process was clearly demonstrated in several ways, notably by the renewed formation of dentin in the incisors of guinea-pigs, the deposition of a homogeneous matrix by the periosteal layer of cells, and finally, the formation of osteoid and osseous trabeculae in the *Gerüstmark* at the costochondral junctions. More recently Wolbach demonstrated that the deposition of collagen in the organization of blood clots in the state of absolute scurvy is referable to the administration of vitamin C, and that it doubtless represents the product of fibroblastic secretory activity.

In an endeavor to determine whether these histologic effects were due to vitamin C per se we isolated this vitamin in crystalline form (ascorbic acid) from lemon juice and administered it to scorbutic guinea-pigs. The ascorbic acid was administered orally or parenterally in a dosage of from 3 to 5 mg. a day. The animals were killed at from three to fifteen days. The reparative reactions were studied in microscopic sections of both the incisor teeth and the costochondral junctions. Typical responses, as previously obtained with orange juice and as stated in the foregoing paragraph, were observed. In an animal which had received four parenteral injections of ascorbic acid the deposition of osteoid matrix in the *Gerüstmark* was particularly marked. Areas of newly formed cartilage were frequently found within the recently deposited collagen matrix at the costochondral junction. In a guinea-pig killed at about two weeks the newly formed osteoid trabeculae at the *Gerüstmark* were found ossified with evidence of progressive bone resorption. This study clearly demonstrates that the cessation in the deposition of intercellular substance in experimental scurvy is due to the lack of ascorbic acid, a relatively simple chemical substance, having as its empirical formula  $C_6H_8O_6$ .

HETEROTOPIC FORMATION OF TEETH. C. B. HUGGINS, H. R. McCARROLL (by invitation) and A. A. DAHLBERG (by invitation), The University of Chicago.

Using a method that we have already described (Huggins, C. B.; McCarroll, H. R., and Dahlberg, A. A.: *Proc. Soc. Exper. Biol. & Med.* **31**:525, 1934), we found it possible to induce the formation of enamel, dentin and cementum in the abdominal fascia of puppies, by autogenous transplantation of the soft tissues of the developing germ of the permanent canine tooth.

The results may be summarized as follows: Transplantation of the ameloblast layer with its subjacent capillary bed and connective tissue, in a "normal" relationship to the pulp, leads to new formation of enamel and dentin, with persistence of the cylindric character of the ameloblasts. But if the approximately normal

relationship of these layers is not maintained, the cylindric form of the ameloblasts does not persist, the epithelium survives in a stratified squamous form, and no enamel is produced. Thus transplantation of the ameloblast layer alone leads to islands and cords of stratified squamous epithelium, at times with keratohyaline pear formation, but without new formation of enamel.

Transplantation of the odontoblast layer of the pulp leads consistently to dentin formation in eleven or more days.

Hertwig's sheath was successfully transplanted in a number of instances.

**RELATION OF VIOSTEROL AND PARATHORMONE IN PUPPIES ON CONTROLLED DIETS.** G. R. SHARPLESS (introduced by F. W. HARTMAN), Henry Ford Hospital, Detroit.

Three groups of growing pups were fed modified Cowgill diets (*J. Biol. Chem.* **56**:725, 1923) and treated with parathormone. Group 1 received a diet low in vitamin D with a calcium content of 0.33 per cent and a ratio of calcium to phosphorus of 1:1.8; group 2 received the same diet and two drops of viosterol (2,500 D) per dog daily; group 3 received the same diet as group 1 except that no calcium was added.

Group 1 tolerated parathormone well, receiving 2,200 units of parathormone in a period of four months and gaining in weight from 3.32 to 6.02 kilograms. Group 2 did not acquire "immunity" to parathormone, received an average of only 630 units and gained 0.5 kilogram in weight in three and one-half months. Group 3 quickly acquired "immunity" to parathormone, received 2,150 units in three months and gained weight as fast as the controls on the same diet.

At autopsy, group 1 showed absence of subcutaneous and omental fat, but no calcium deposits were visible in gross. Group 2 showed metastatic calcification in the stomach, heart, kidneys and thyroid gland. An x-ray picture showed suggestive cyst formation in the femur of one dog in group 2, but no change in either of the other groups. Group 3 appeared perfectly normal and well nourished. In none of the groups were the bones deformed or extremely soft.

**PARATHYREOTROPIC ACTION OF ANTERIOR LOBE OF THE PITUITARY GLAND; HISTOLOGIC EVIDENCE IN THE RABBIT.** SAUL HERTZ and ALFRED KRANES (introduced by JAMES H. MEANS), Massachusetts General Hospital, Boston.

On the basis of a preliminary observation that emulsions of fresh anterior lobe of the pituitary gland gave rise to an enlargement and increased vascularity of the parathyroid glands of the rabbit, a more complete histologic study was undertaken. In a series of seven experiments the parathyroid glands of forty-eight rabbits subjected to injections of anterior lobe extract and of pregnancy urine as well as of several proprietary preparations from these sources were grossly and microscopically compared with those of seventeen reference control animals. The latter either received no injections or were given injections of heated pituitary, normal urine or emulsion of fresh brain substance.

The animals which had been treated with anterior lobe preparations and extracts of pregnancy urine exhibited parathyroid glands which were grossly larger and more vascular than those of the control group. They showed histologic changes consistent with hypertrophy and hyperplasia. The criteria for the latter described by Erdheim in reference to the rat were applied.

**THE PROSTATE AND SEMINAL VESICLES AS QUANTITATIVE INDICATORS OF THE MALE SEX HORMONE.** ROBERT A. MOORE, Cornell University.

The male sex hormone content of any extract may be measured by the increase in the size of the comb in the capon or by the height of the prostatic and vesicular epithelium of the castrated rat. In confirmation of the investigation carried out by Hansen, I have found that the height of the epithelium of the adult male rat, castrated and treated by injection for ten days and killed on the eleventh day, shows a direct relationship to the amount of hormone injected.

**CORRELATION OF ANTEMORTEM AND POSTMORTEM BACTERIOLOGY.** CASPAR G. BURN (introduced by RAYMOND HUSSEY), Yale University.

In a bacteriologic study of eighty-three cases in which blood cultures were made both before and after death, agreement was found in fifty and disagreement in thirty-three. Among the cases in which there was disagreement there were five in which contamination of the blood cultures was the result of technical difficulties in the collection of the material. Among the remaining twenty-eight there were nineteen in which the last clinical culture was taken three days or longer before death, and nine in which the interval before death was less than forty-eight hours. In contrast, among instances in which the antemortem and postmortem cultures were in agreement there were 31 in which the clinical cultures were taken within forty-eight hours of death. Bacteriologic studies of the organs in fifty-seven of the eighty-three cases demonstrated bacteria other than the kind isolated from the blood stream. Postmortem blood cultures agree in a significantly large number of instances provided the antemortem cultures are obtained shortly before death. Organs at necropsies contain pathogenic bacteria other than the kinds isolated from the blood stream.

**THE INFLUENCE OF COPPER IN THE DIET ON THE RESISTANCE OF ALBINO RATS TO TRYPANOSOMAL INFECTIONS.** DAVID PERLA (introduced by DAVID MARINE), Montefiore Hospital, New York.

In previous work it was found that the addition of small amounts of copper to an adequate diet for rats protected a large percentage of the rats from *Bartonella muris* anemia following splenectomy. Further studies were undertaken to determine whether copper added to the diet would raise the natural resistance of adult albino rats to trypanosomal infections. The addition of copper (0.1 mg. per rat per day) or iron (1 mg. per day) or both to an adequate diet during a period of ten days prior to an induced infection with *Trypanosoma Lewisi* raised the natural resistance of the rats to this infection. The infection was completely aborted in almost 50 per cent. Lead salts added to the diet had no beneficial effect.

The addition of copper in amounts of from 0.2 to 0.4 mg. per rat per day to an adequate diet during a period of ten days prior to an induced infection with *Trypanosoma equiperdum*, a fatal infection in rats, strikingly raised the natural resistance of the rats to this disease. Adult albino rats infected with overwhelming doses of *T. equiperdum*, if previously fed copper, survived slightly longer than the controls. Adult albino rats fed a diet supplemented with 0.4 mg. of copper per rat per day during a period of ten days prior to the injection of 10,000 trypanosomes (*T. equiperdum*) per rat developed only an abortive infection and 60 per cent recovered, whereas all the controls died. When the rats were infected with 2,000 trypanosomes per animal, all the copper-fed rats developed abortive infections and recovered, but 62 per cent of the controls died.

It is concluded that the species susceptibility of rats to trypanosomal infections may be markedly altered by supplementing their diet with copper prior to infection.

**CELLULAR CHANGES IN THE SPLEEN IN PERNICIOUS ANEMIA.** RAPHAEL ISAACS, University of Michigan.

In films of serum suspensions of the spleens of patients with pernicious anemia there is a relative and an absolute increase in the number of elongated bipolar cells with oval nuclei (9 by 6 microns) having the characteristics of "connective tissue cells." The isolated cells have two long cytoplasmic processes, occasionally with bifurcated ends. These are from 90 to 105 or more microns in length and average 1.5 microns in width. A similar increase in the number of these cells is noted in fetal spleens and, to a lesser degree, in the spleens of patients who have had considerable irradiation with roentgen rays. The increase in the number of these cells appears to be characteristic of pernicious anemia, the cells constituting from 13 to 25 per cent of all nucleated cells as compared with less than 6 per cent in other conditions. Data on the number of these cells in relapse and in remission of pernicious anemia and in certain other diseases are given.

## THE NATURE OF THE RESPONSE TO INFECTION IN DYSCRASIA OF BONE MARROW.

FRANK H. BETHELL (introduced by CYRUS C. STURGIS), University of Michigan.

During the course of an infection alterations in the maturity of the neutrophils as evidenced by their nuclear form are dependent on the severity of the infection and the functional state of the bone marrow prior to the acute illness. Cytoplasmic changes in the neutrophils and in particular the appearance of basophilic or "toxic" granules in Wright-stained preparations are related solely to the infective process, being quite independent of the nature of the bone marrow response. That such a modification of the staining characteristics of the neutrophil cytoplasm is not a merely local phenomenon dependent on exposure to a site of inflammation is attested by the evidence gained from successive estimations of the total neutrophil count, the nuclear shift and the incidence of basophilic granulation in the immature and adult classes. During the period of exacerbation basophilic granulation is increasingly prevalent in the immature neutrophils most recently released into the circulation.

In cases of abnormal bone marrow response such studies provide an insight into the nature of the myeloid dysfunction. The examples of atypical response to infection which I shall report comprise pernicious anemia in an aregenerative phase and at the beginning of a therapeutically induced remission, postarsphenamine aplastic anemia, agranulocytic angina and aleukemic lymphatic leukemia.

FUNDAMENTAL BONE MARROW REACTIONS: II. THE EFFECT OF SMALL DOSES OF X-RAYS AND RADIUM ON MYELOPOIESIS IN THE PIGEON AND RABBIT.  
CHARLES A. DOAN and LOWELL A. ERF (by invitation), Ohio State University.

Nakahara in Murphy's laboratory believed that he was able with very small doses of x-rays to produce a primary stimulation of cells in the germinal centers of lymph nodes as reflected by an increase in the number of mitotic figures. However, he noted no change in the bone marrow elements.

In a series of experiments in rabbits, Sabin, Doan and Forkner, using monthly injections of radium chloride and mesothorium, 5 and 7 micrograms respectively, produced definite pathologic changes but, despite extensive foci of necrosis and fibrosis in the bone marrow, granulopoiesis continued to maintain the neutrophils at an approximately normal level through the experimental period of from nine to sixteen months. No absolute neutrophilic leukocytosis occurred as a result of the injection of these radioactive materials.

In pigeons, in which myelopoiesis and erythropoiesis are sharply separated and aplasia and hyperplasia coexist in different areas, we have used doses of x-rays of high voltage ranging from 19 roentgens ( $\frac{1}{40}$  skin erythema dose) to 790 roentgens (1 skin erythema dose), both singly and in series, and have in no instance found any increase in mitosis of myeloid elements in the absence of definite myelocytic destruction. Biopsies were made in each instance before irradiation, and any change in the gross or microscopic evidence of myelopoiesis was always found to be directly proportional to the extent of specific myelocytic nuclear degeneration and karyolysis. No absolute increase in circulating granulocytes resulted in any of these animals.

We therefore conclude that it is probably impossible to secure primary myelopoietic stimulation by the use of even the smallest, so-called, therapeutic doses of x-rays.

EFFECT OF THE HOST CONSTITUTION ON THE LYMPHOID CELLS OF TRANSMISSIBLE MOUSE LEUKEMIA. J. S. POTTER and JOSEPH VICTOR (introduced by JAMES W. JOBLING), The Carnegie Institution of Washington and Columbia University.

Two lines (I and M-Liver) of cells of transmissible lymphatic leukemia which have been carried in genetically homogeneous mice of strain C58 yield similar gross and microscopic pictures and kill hosts within similar periods of time. When

either of these lines is carried through mice of the highly inbred Storrs-Little strain, the picture presented by the infiltration is the same for the first seventy-two hours as that found in strain C58. After this period the lesions may increase in size and number, eventually causing death of the host, or they may regress, terminating in recovery of the host.

The cells of both these lines show a reduced mean number of mitochondria when growing in hosts of the Storrs-Little strain. In line M-Liver the mode as well as the mean is changed; in line I only the mean is changed. Cell size and other morphologic traits remain the same in hosts of each strain.

The changes in the number of mitochondria are coincident with changes in the glycolytic rate in these transmission lines, suggesting a direct relationship. When line cells are returned to hosts of strain C58 all functions return to the level observed before the passage through the Storrs-Little strain.

THE METABOLIC EFFECT OF THE HOST CONSTITUTION ON THE LYMPHOID CELLS OF TRANSMISSIBLE MOUSE LEUKEMIA. JOSEPH VICTOR and JAMES S. POTTER (introduced by JAMES W. JOBLING), Columbia University and The Carnegie Institution of Washington.

The coefficients of variability for repeated determinations of the oxygen consumption and the aerobic and anaerobic glycolysis of a given line of leukemic lymphocytes carried by host mice of strain C58 vary from 3 to 15 per cent. This variability, smaller than any reported in the literature for transplantable tumor, may be attributed in part to the genetic uniformity of mice of strain C58. Normal lymphocytes of this strain do not differ metabolically from those of another genetically uniform strain of mice, Storrs-Little. When leukemic cells of line I were transferred from strain C58 to hosts of the Storrs-Little strain, the oxygen consumption increased, and the aerobic and anaerobic glycolysis diminished; leukemic cells of line M-liver gave a similar reduction in glycolysis, but no change in oxygen consumption. When these two lines of cells were transferred from a Storrs-Little donor to hosts of strain C58, the metabolic rates returned to their original levels. These results indicate that the genetic constitution of the host affects the metabolism of leukemic cells, and that different lines of cells may be differentially affected by the same host. Furthermore, the metabolic effect of a host on leukemic cells is temporary and present only when the cells are in that host.

CHANGES IN VIRULENCE AND SIZE IN LINES OF LEUKEMIC LYMPHOCYTES CARRIED BY MICE. MAURICE N. RICHTER and E. C. MACDOSELL (by invitation), Columbia University and The Carnegie Institution of Washington.

Changes in the interval between inoculation and death are frequently observed during the course of successive transfers of a line of leukemic lymphocytes. When uniform technic and genetically and ontogenetically homogeneous hosts are used, these changes indicate changes in the cell line. The changes usually but not always shorten this interval. They may occur at any time and at any rate from abrupt changes within a single transfer to very slow ones extending over a long series of transfers. Stable periods with constant averages and degrees of variability occur at any time after the first three transfers.

Cytologic studies have been made on specially prepared material covering the periods of more rapid change in the aforementioned interval. The observations include the cell size, the rate of division and the ratio of the nucleolus to the nucleus. In periods when this interval is stable cytologic traits are stable. Cytologic changes are found to occur during periods of change in this interval and indeed may be detected in advance of the latter change. Cell traits may alter gradually over a series of transfers that include a sudden break in the length of this interval. Cell changes are antecedent to changes in this interval, but threshold phenomena in certain cases may lead to abrupt changes in the interval.

**A NEOPLASTIC DISEASE WITH NUCLEAR INCLUSIONS OCCURRING IN THE LEOPARD FROG.** BALDUIN LUCKÉ, The University of Pennsylvania.

In about 2 per cent of leopard frogs (*Rana pipiens*) there has been observed a neoplastic disease which manifests itself by the occurrence of unencapsulated whitish tumors in the kidneys. In the majority of the cases these tumors have the histologic appearance of adenocarcinoma.

Two hundred and four frogs with such tumors have been studied. The disease occurs in the two sexes and in the two kidneys with approximately the same frequency. The tumors are commonly multiple and in about one half of the cases occur bilaterally. They vary in size from a minute nodule to a mass several times larger than a normal kidney. While the new growths are locally destructive and infiltrative, they rarely metastasize.

In the majority of the tumors there are present characteristic nuclear changes, namely, a condensation of chromatin at the periphery of the nucleus and very numerous and prominent acidophilic inclusion bodies lying within nuclei which otherwise appear almost empty. The inclusions are of the same general appearance as those of herpes, chickenpox and certain other virus diseases. They occur only in the epithelial cells of the tumors. Such inclusion bodies are currently regarded as presumptive evidence of the activity of a virus; however, proof of the existence of a virus and of its relation to the tumor cannot be furnished.

Material from twenty-four different tumors has been transplanted by various methods into several regions of four hundred and seventy-eight frogs. In a relatively small number of animals the transplants have survived for upward of three months and exhibited some evidence of growth, but no massive tumors have developed at the sites of inoculation. However, in a number of the animals large tumors did arise in the kidneys; this may or may not be evidence of transmission. It should be pointed out that if the causal agent of this neoplastic disease is a virus the virus would be expected to localize and produce, under experimental conditions, lesions in that tissue (in this case, the kidney) in which the spontaneous growths occur. Further experiments are necessary to establish the nature of the tumors. They appear to be promising material for various studies on new growths.

**THE PROLIFERATIVE REACTION OF GUINEA-PIG SKIN TO SULPHYDRYL AND ITS RELATION TO NEOPLASIA.** STANLEY P. REIMANN and ETHEL RAHE HANKELE (by invitation), The Lankenau Hospital Research Institute, Philadelphia.

This article will appear in full in a later issue of the ARCHIVES.

**AN APPARENTLY HEREDITARY FORM OF SPLENOMEGLY WITH CIRRHOSIS OF THE LIVER IN THE RABBIT.** HARRY S. N. GREENE, PAUL D. ROSAHN and C. K. HU (introduced by W. H. BROWN), The Rockefeller Institute for Medical Research, New York.

A pathologic complex consisting of diffuse fibrosis of the spleen and portal cirrhosis of the liver has been found in a line of inbred rabbits with a frequency suggesting that it is hereditary. The condition bears a marked resemblance to the splenic anemia of man and is being studied with a view to determining its relationship to this symptom complex and its pathogenesis.

**THE EFFECT OF OXYGEN IN PREVENTION OF HEPATIC NECROSIS PRODUCED BY VOLATILE ANESTHETICS.** S. GOLDSCHMIDT, I. S. RAVDIN and B. LUCKÉ, The University of Pennsylvania.

A comparison has been made of the relative incidence of hepatic necrosis in dogs anesthetized in a semiclosed system when the anesthetic was volatilized with air, and when it was volatilized with oxygen. The data show that the use of oxygen with either divinyl ether or chloroform is a potent factor in the reduction of postanesthetic necrosis of the liver.

When divinyl ether was used as the anesthetic by any method, necrosis of the liver was not observed with any degree of regularity until the anesthesia had been maintained for a period of three hours. The incidence of postanesthetic necrosis after three hours of anesthesia in a semiclosed system was nearly twice as high when the anesthetic was volatilized with air as with oxygen.

Chloroform anesthesia, on the other hand, resulted in a high incidence of hepatic necroses in dogs after one hour of anesthesia. The incidence of necrosis of the liver following one hour of chloroform anesthesia in a semiclosed system was approximately ten times as great when the anesthetic was volatilized with air as with oxygen.

Hepatic degeneration has been produced in the dog following three hours of ether anesthesia when the anesthetic was volatilized with less than the atmospheric pressure of oxygen, i. e., oxygen 15 per cent and nitrogen 85 per cent.

Data demonstrating the efficacy of oxygen during anesthesia will be presented, and the general implications of our findings will be discussed.

**SERUM PHOSPHATASE IN EXPERIMENTAL INSUFFICIENCY OF THE LIVER.** F. W. HARTMAN and VICTOR SCHELLING (by invitation), Henry Ford Hospital, Detroit.

In the course of experimental insufficiency of the liver produced with a modified Eck fistula and x-rays of high voltage, renal stones as noted by other observers were found in about 50 per cent of the animals that lived more than six months after operation. This observation led us to determine the phosphatase in normal animals and in animals with hepatic insufficiency produced by Eck's fistula, Eck's fistula plus irradiation with x-rays of high voltage and carbon tetrachloride. All of these groups showed a marked increase in serum phosphatase ranging from five to ten times the normal. Further there is a close parallel between the increase in phosphatase and the function of the liver as determined by cholesterol partition, bilirubin and bromsulphalein.

**INTOXICATIONS OF DOGS WITH BILIARY FISTULA.** WILLIAM B. HAWKINS, The University of Rochester, Rochester, N. Y.

Three types of dogs with biliary fistula have been studied over long periods with observations as to the different states of intoxication that develop following deprivation of the intestinal tract of bile.

Such animals are subject to gastro-intestinal upsets if not fed a suitable diet.

In dogs with open infected fistula and obstructed infected closed fistula the bones are progressively decalcified, with resulting multiple fractures. Liver added to the diet prevents or delays this change. Dogs with renal fistula may live for long periods deprived of bile without this decalcification.

Fistulous dogs deprived of bile show purpuric tendencies with hemorrhage from the gastro-intestinal tract or trivial wounds. Their blood reveals normal fibrinogen and normal platelets, and the blood will clot in a test tube after some delay. Serum added to recalcified plasma causes the formation of the clot in normal time. Whole bile or bile salts given by mouth apparently restore conditions to normal with a disappearance of the bleeding tendency.

Fifty cubic centimeters of whole dog or ox bile by mouth will maintain a fistulous dog in a healthy condition for long periods of time.

Combination biliary and Eck fistulas are not well tolerated by dogs.

**THE OVERPRODUCTION OF BILIARY PIGMENT IN SPLENECTOMIZED DOGS WITH BILIARY FISTULA.** RALPH E. KNUTTI, The University of Rochester, Rochester, N. Y.

In splenectomized dogs with biliary fistula periods of marked production of bilirubin occur. Associated with such periods is a massive destruction of red blood cells. The amount of biliary pigment produced at these times is greatly in excess

of the amount that can be accounted for by the bilirubin equivalent of destroyed hemoglobin, although there are very rapid destruction and rebuilding of this substance. The occurrence of these periods has been found to be related to the presence in the red blood cells of bodies indistinguishable from those of *Bartonella canis*. Modification and disappearance of such periods have been brought about by the oral administration of an extract of spleen. An attempt to explain the disparity of the ratio of bilirubin to hemoglobin has been made.

**BILIARY CHOLESTEROL: FLUCTUATIONS DUE TO DIETARY FACTORS, BILIARY SALT, INJURY TO THE LIVER and HEMOLYSIS.** ANGUS WRIGHT (introduced by GEORGE H. WHIPPLE), The University of Rochester, Rochester, N. Y.

Observations were made on dogs with sterile closed biliary fistulas. The method used in determining the values of biliary cholesterol was quantitatively accurate. The effect of various factors on the excretion of biliary cholesterol was determined after control levels had been established on a basal diet. Under uniform dietary conditions dogs eliminate fairly constant amounts of biliary cholesterol. Diets rich in cholesterol raise the cholesterol output in the bile. Biliary salt alone raises the biliary cholesterol as much as or more than a diet rich in cholesterol. Biliary salt plus a cholesterol-rich diet gives the maximal output of biliary cholesterol. Hepatic injury decreases the elimination of both cholesterol and biliary salt in the bile. Destruction of red blood cells fails to increase the output of biliary cholesterol.

**COMPARISON OF THE RESULTS OF THE WASSERMANN TEST IN TWO LABORATORIES.** DAVID L. BELDING, The Massachusetts Memorial Hospitals, Boston.

A statistical study of the Wassermann test on 10,000 duplicate samples of serums of known reaction strength from treated syphilitic patients was made in two laboratories. Of the serums 30 per cent gave reactions of less than 1 unit; 20 per cent, above 4 units, and 50 per cent, between 1 and 4 units.

If these serums had been tested in one laboratory, 17 per cent of the positive reactions would have been missed. Lack of agreement was almost entirely confined to the borderline group of serums with reactions of from 1 to 4 units, in which there was 35 per cent disagreement. The daily variation in laboratory technic was the chief cause of this disagreement. The daily variations from an arbitrary mean of 100 gave standard deviations of 34.9 and 23.7, respectively, for the two laboratories.

**THE EFFECT OF IODINE AND VITAMIN B<sub>1</sub> ON THE THYROID, SUPRARENAL AND HYPOPHYSEAL GLANDS OF THE RAT.** M. D. CARPENTER and G. R. SHARPLESS (introduced by F. W. HARTMAN), Henry Ford Hospital, Detroit.

A study has been made of the effect of iodine and of vitamin B<sub>1</sub> on the weight of the thyroid, the suprarenal and the hypophyseal glands, the structure of the thyroid gland and the iodine content of this gland. The rat was used, and studies were made at the end of 41, 82 and 123 days of the experimental diet.

The results calculated as weight of gland per hundred grams of body weight show a definite enlargement of all three glands during deficiency of vitamin B<sub>1</sub>. The percental enlargement is much greater for the thyroid than for the suprarenal or the hypophyseal gland. There seems to be no consistent correlation between the size of either the suprarenal or the hypophyseal gland and the iodine intake, but the thyroid gland is definitely smaller in the animals with the higher iodine intake.

Iodine determinations on the thyroid gland show that an increased iodine intake increases the iodine content of this gland, but the percental iodine content of the gland does not seem to be closely correlated with the size of the gland.

Microscopically, definite extensive hyperplasia of the thyroid gland was evident only in those rats receiving a diet low in iodine and optimal in vitamin B<sub>1</sub>. There is some evidence of accumulation of dense colloid in those receiving iodine during deficiency of vitamin B<sub>1</sub>.

**THE IODINE CONTENT OF THE BLOOD IN DISEASES OTHER THAN THOSE OF THE THYROID GLAND.** GEORGE M. CURTIS, Ohio State University.

The majority of hospital patients with diseases other than those of the thyroid gland present a normal content of iodine in the blood. Among the diseases investigated were Hodgkin's disease, tuberculosis, cancer, fractures, chronic osteomyelitis and functional disorders. Likewise, the majority of ambulatory patients with such diseases present a normal content of iodine in the blood. There are, however, certain exceptions. In acute severe infections—for example, septicemia—the level of the blood iodine is elevated. It is likewise elevated in lymphatic leukemia. It rises immediately following a major surgical operation for disease other than that of the thyroid gland. It is elevated in certain forms of heart disease and in hypertension. Since the level of the blood iodine is a measure of the activity of the thyroid gland, it is possible to use it in evaluating the thyroid component in diseases other than those of the thyroid gland.

**CHANGES IN THE CENTRAL NERVOUS SYSTEM RESULTING FROM CONVULSIONS DUE TO HYPERINSULINISM.** DAVID M. GRAYZEL (introduced by RAYMOND HUSSEY), Yale University.

Repeated convulsions for varying periods of time were induced artificially by the production of hyperinsulinism. The convulsions were produced by the intravenous injection of insulin into rabbits that previously had been made to fast for eighteen hours. The dose of insulin required to produce an attack varied considerably (from 2 to 12 units) for different animals. The attacks occurred within from two to six hours after the injection of the insulin. The convulsions were allowed to continue for periods of time ranging from three minutes to several hours. They were then terminated by the intravenous injection of 10 cc. of 50 per cent dextrose. Whenever possible the experiments were conducted over a period of three months. At the close of the experiments the animals were killed with ether and examined. Blocks of tissue were taken from various parts of the brain, fixed in 95 per cent alcohol and stained with toluidine blue by the Nissl method. The lesions were arbitrarily graded from negative to 4 plus depending on the severity.

From the results it was apparent that animals which had not been convulsed, or only slightly, showed either minimal or no cerebral changes regardless of the number of injections of insulin that they had received. On the other hand, even one convolution, if prolonged and severe enough, produced definite lesions in the central nervous system, of the necrobiotic type. Furthermore, the more prolonged and the more severe the convulsions, the more extensive were the lesions.

**GENETIC VARIATIONS IN THE PERIOD OF GESTATION OF THE RABBIT.** PAUL D. ROSAHLN, HARRY S. N. GREENE and C. K. Hu (introduced by W. H. BROWN); The Rockefeller Institute for Medical Research, New York.

During the past few years this laboratory has maintained a colony comprising approximately one thousand five hundred standard-bred and hybrid rabbits. The data accumulated include accurate records of the date of mating and the date of birth of the offspring of all animals raised in the colony. This information has been submitted to a statistical analysis, with the general purpose of determining the period of gestation of the rabbit, and the particular purpose of determining whether the breed of rabbit has any influence on the duration of pregnancy. The mean gestation period has been calculated for several standard-bred lines, and it has been found that statistically significant differences exist between different breeds. Moreover, the variation between breeds was significantly greater than the variation within the breeds. These findings indicate that the genetic factors which are responsible for differences in breed significantly influence the period of gestation of the rabbit.

HEREDITARY HYDROCEPHALUS IN THE RABBIT. HARRY S. N. GREENE, C. K. HU and PAUL D. ROSAHLN (introduced by L. PEARCE), The Rockefeller Institute for Medical Research, New York.

The occurrence of hydrocephalus in certain hybrid lines of the rabbits in our breeding colony has long attracted our attention. The condition is manifested early in life in the young of male and female transmitters.

It appears that this abnormality has more than one form. The primary variations responsible for the difference in form have not been determined. In some instances the hydrocephalus is internal and in other cases it is external. Furthermore, it may or may not be associated with deficient or retarded calcification of the membranous bones of the skull.

Most of the animals manifesting the abnormality die early in life. However, recent experiments indicate that a therapeutic measure for hastening the calcification of the calvarium is capable of checking the progress of the hydrocephalus, and life may be prolonged and possibly saved. Further investigations are in progress.

It is possible that such a therapeutic measure for hastening calcification of the calvarium, when carried out early enough, may prove to be of practical use in certain cases of hydrocephalus in man.

METASTASIS OF A SQUAMOUS CELL CARCINOMA FROM THE WRIST TO THE AXILLA WITHOUT DEMONSTRABLE INTERVENING GROWTH. ESMOND R. LONG, The Henry Phipps Institute, Philadelphia.

Since opinion is divided as to whether lymphatic spread from a primary carcinoma to distant secondary lymph nodes occurs by lymphatic embolism or by continuous lymphatic growth a study was made of the entire amputated arm from a man with a carcinoma of the wrist.

The patient, 59 years of age, entered the Billings Hospital of the University of Chicago with a squamous cell carcinoma of the right wrist. Enlarged glands were palpable in the right axilla. The limb was amputated in the middle third of the upper part of the arm, and the lymph nodes in the axilla were removed *en masse* by block resection. Several of these contained carcinomatous nodules. The patient made an uneventful recovery. Two years later he was well, with no evidence of recurrence.

Sections were taken from serial blocks from the wrist to the upper end of the lower third of the amputated arm. The arm was first fixed in Zenker's solution by injection through the main arteries, and sawed into twenty-seven consecutive blocks from 1 to 1.5 cm. thick. The bones were then removed and the blocks embedded in pyroxylin (celloidin). Several representative sections from each block were stained with hematoxylin and eosin. Blocks 1 and 2, from the wrist, contained carcinoma with definite extension into the lymphatics. In block 3 a few cells thought to be from the carcinoma were seen. Considerable granulation tissue was present at the site corresponding to the carcinoma in the previous blocks. In blocks 4 and 5 traces of granulation tissue persisted, but no carcinoma cells were seen. In block 7, the first block with large muscle trunks, a scattered lymphangitis was discovered, several lymphatics being plugged with polymorphonuclear leukocytes. The reason for this was apparent in most of the succeeding blocks, in the presence of many partially encysted larvae of *Trichinella*. A severe myositis due to these parasites was found in the muscles of the upper part of the arm. The lymph nodes of the elbow were in view in blocks 22 to 24. Many sections were cut from these blocks and no carcinoma was seen. Sections from all of the bones removed from the blocks of the forearm were studied and no carcinoma was found. Therefore, as no carcinoma was found above the wrist, it was concluded that the metastases in the axilla were due to lymphatic embolism, and that this study had demonstrated that metastasis of carcinoma may occur without continuous growth, whether this is the usual method or not.

A STUDY OF MUSCLE TEMPERATURE DURING BACTERIAL CHILL. A. J. NEDZEL  
(introduced by WILLIAM F. PETERSEN), University of Illinois.

By direct measurements of the temperature in the abdominal aorta, rectum and skeletal muscles, the temperature response to the intravenous injection of *Bacillus coli* has been followed. In the majority of experiments the temperature rose slowly in the abdominal aorta and the rectum while in the muscles there was a definite decrease in temperature. The conclusion is reached that the elevation of temperature in the circulating blood does not depend on the heat produced in the muscles during the period of chill.

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BUFFALO PATHOLOGICAL SOCIETY

*Regular Meeting, Feb. 23, 1934*

KORNEL TERPLAN, *President, in the Chair*

OXYURIASIS OF THE APPENDIX. MARGARET WARWICK.

During the past five years at the Millard Fillmore Hospital 3,196 appendixes were removed. In 1,911 of these the lumens showed contents so preserved that examination for oxyurids could be satisfactorily made. Parasites were found in 35, or 1.8 per cent. By years the incidence has increased from 1 per cent in 1929 to 2 per cent in 1934. The ages of the patients varied from 7 to 34 years, with an average of 17.8 years. That appendical oxyuriasis is not entirely an infestation of childhood was shown by the fact that 51 per cent of the cases were found in patients over 16 years of age, 31 per cent in patients over 20 years and 8.5 per cent in patients over 30 years.

In 60 per cent of the cases in which oxyurids were found the appendix alone was removed; in 40 per cent appendectomy was incidental to operations on other tissues; one appendix was contained in a hernial sac. The symptoms for which appendectomy was done varied from a light attack of abdominal pain lasting over a long period of time to acute attacks of from one to two days' duration. Leukocyte counts were done in fifteen cases. In 53 per cent of these the count showed an increase of from 10,000 to 28,000. For the most part the temperature did not exceed 99 F.; in six cases, or 17 per cent, the temperature reached 100 F.; in two others, in which there were acute appendicitis and pyelitis, the temperature was higher.

In only one case did the appendix show signs of acute inflammation; the parasite was in the lumen surrounded by pus. A few appendixes showed indentations of the mucosa, but no erosions or ulceration. In three, the parasite had penetrated into the mucosal coat. There was no surrounding cellular reaction. Gordon concluded that such a penetration occurred after removal of the appendix. Two of the parasites were surrounded by red blood cells in the lumen. The location of the hemorrhage, however, between the mucosa and a central mass of fecal material suggested a traumatic origin associated with removal. In most cases the worms lay in the lumen, either alone or in a mass of fecal material.

An attempt at correlation of the clinical symptoms and the position of the worm within the lumen showed that the acute attacks of pain were usually associated with the presence of unusually large or numerous parasites in empty lumens. This suggests that, as Aschoff thought, the movements of the oxyurids might cause painful contraction of the appendical wall which could produce symptoms resembling acute appendicitis. It is difficult, however, to believe that contraction could have a relation to increase in the leukocyte count.

The presence of the oxyurids in appendixes is usually an incidental finding. The worms are probably never the cause of appendical inflammation. They may produce painful muscular contractions which invoke symptoms simulating acute appendicitis.

**LIPOID HISTIOCYTOSIS WITH TUMOR-LIKE INFILTRATION OF THE SUBCUTANEOUS STRUCTURES OF THE TRUNK.** K. TERPLAN, R. S. HUBBARD, C. S. RYERSON, W. J. ROSE AND S. L. VAUGHAN.

A white woman, 35 years of age, was admitted to the Buffalo General Hospital, Sept. 12, 1933, complaining of polydypsia, polyuria and headache of about five years' duration. For two years she had noticed a mass in the right breast, which increased progressively in size, and rapidly growing masses in the neck, left breast and abdomen. There was no exophthalmos. Repeated cholesterol determinations on the blood were within normal limits. Roentgen films of the skeleton were negative. The clinical impression was: Hodgkin's disease or carcinoma of the breasts. The patient died Dec. 9, 1933.

Biopsy of a specimen taken from the mass in the left breast showed no evidence of neoplasm. The picture pointed to a severe metabolic disturbance of the subcutaneous structures with unusual storage of lipoid.

Hematologic examination disclosed a normocytic hyperchromic type of anemia without signs of regenerative activity. Later examinations revealed a progressive anemia with evidences of marked erythropoiesis, especially an erythroblastic crisis; the total leukocyte count fell, with a distinct shift to the left in the neutrophilic nuclear configuration.

Various portions of the swelling removed from the left breast were studied by chemical methods. Attention was centered on the lipoid constituents, and the composition of the stroma was ignored. The lipoids were apparently wholly contained in an acetone extract, for although a second portion soluble in alcohol but insoluble in acetone was obtained, it was also wholly soluble in water but insoluble in ethyl ether and petroleum ether. The relationship between the nitrogen and the phosphorus content of this mixture did not suggest that an alcohol-soluble phospholipid was present.

The material soluble in acetone was also wholly soluble in ethyl ether and practically completely soluble in petroleum ether. It consisted largely of cholesterol ester, together with a small amount of free cholesterol and a trace of phospholipid. There was also present some neutral fat, as well as, apparently, some free fatty acid. The evidence in favor of free fatty acid was: 1. A definite titration value was obtained when a benzene solution prepared from the acetone extract was titrated with alkali. 2. After exact or partial neutralization with alkali followed by extraction and determination of the fatty acid the amount recovered was proportional to the amount of alkali initially added. 3. After cholesterol ester had been removed by crystallization from methyl alcohol a small crop of crystals and amorphous globules was obtained by treatment with ethyl alcohol. These crystals had a very low melting point, did not rotate polarized light, were acid in reaction and stained light yellow with Sudan III.

The cholesterol ester, together with a part of the free cholesterol, was obtained in crystalline form by treatment with methyl alcohol. The proportion between the total weight of the crystalline material and that of the cholesterol in the ester form present corresponded with the theoretical figures for cholesterol stearate or cholesterol oleate. By purification with digitonin and repeated purification, first from ether and then from methyl alcohol, a preparation melting sharply between 41 and 42 C. was obtained. Synthetic cholesterol oleate, according to Hurthel, melts at 42 C.

Post mortem there were found: extreme pallor of the entire integument; seborrheic dermatitis limited to the scalp; marked caries of the teeth; nodular lumpy and diffuse lipoid masses of both lateral thoracic walls extending into the axillae, both cervical regions and the submental areas, almost the entire back, and about the upper half of the abdominal wall (deepest depth, 8 cm.); lipoid infiltration of muscles; moderate swelling of cervical lymph nodes with localized caseation; lipoid infiltration in the mediastinum and in the retroperitoneal and pelvic fat tissues; localized lipoid infiltration of the dura mater with thrombosis of several sinuses, and hemorrhagic malacia in the brain; distinct lipoid deposits in the fat marrow of the long bones, and to a slight extent in the short bones (skull not

involved); extensive thrombosis of the iliac and hypogastric veins extending into the superior vena cava; complete thrombosis of the splenic vein, with extensive anemic infarction; thin whitish shreds on the entire mesentery suggestive of organized fibrinous exudate; atrophy of the thyroid gland (6.5 Gm.); normal lipoid-rich suprarenals; very slight atheromatosis.

Histologic examination of subcutaneous tissue pointed to a gradual replacement of the fat by huge foamy cells stored with double-refracting lipoid, and to a reactive inflammation, in places leading to formation of granulation tissue. With Sudan III, Nile blue sulphate, Weigert-hematoxylin, Lorrain-Smith and Fischler stains, most of the lipoid consisted apparently of cholesterol oleates. Study of the borders of large areas of foamy cells where small fat islets were still preserved suggested the appearance of double-refracting lipoids in structurally normal fat cells. No lipoid storage could be made out in the endothelium or reticulum cells of the spleen, liver or lymph nodes. Of pathogenic interest was the practically complete fibrous replacement of the posterior lobe of the pituitary gland. The histologic changes in the lungs were similar to those found in some cases of the Christian-Schüller syndrome. The bone marrow, along with the lungs, showed huge foamy cells in sections examined. It is interesting to note that at no time was the blood cholesterol increased.

We were able to find only one case reported in which the subcutaneous tissue was involved by massive lipoid deposits. In 1909 under the title of "Multiple Myxo-Cholesto-Lipomata," Proescher and Meredith described the presence of multiple yellowish tumor masses in the subcutaneous tissues of the leg, breasts and abdomen of a 32 year old woman. From chemical analysis it was concluded that the foreign material was abnormally stored cholesterol fatty acid esters, but the malady was regarded as a tumor process. However, the patient had no diabetes insipidus.

NOTE.—It is intended to publish a more detailed description of this case at a later date.

## Book Reviews

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**Bergey's Manual of Determinative Bacteriology: A Key for the Identification of Organisms of the Class Schizomycetes.** By David H. Bergey. Assisted by a committee of the Society of American Bacteriologists. With an index by Robert S. Breed. Fourth edition. Cloth. Price, \$6. Pp. 664. Baltimore: Williams & Wilkins Company, 1934.

Following the preface, the chief divisions of the contents of the recently published fourth edition of Bergey's manual are: (1) introduction; (2) an abridged key of the bacterial families, tribes and genera; (3) a section "How Bacteria Are Named and Identified," by Prof. R. E. Buchanan; (4) the latest bacteriologic code; (5) descriptions of species of bacteria, which comprise the bulk of the book, and (6) the index, prepared by Prof. R. S. Breed.

The introduction contains a slightly revised version of the previously published suggestions for the use of the manual in classifying unknown organisms.

The abridged key, adapted from the "Manual of Microbiology" of Obold and Diehm, summarizes the principal characters of the different genera of bacteria. It should be of considerable aid in the allocation of a particular species.

Professor Buchanan's essay "How Bacteria Are Named and Identified," written especially for this edition, compresses into twelve pages a clear and stimulating discussion of the principles of taxonomy and nomenclature. It contains statements of broad biologic concepts, together with a great deal of practical information on the scientific names of the bacteria. The discussion, on page 18, of the concept of species in relation to R and S types, filtrable stages, dissociation phenomena, variability and possible life cycles of the bacteria is particularly interesting and is good evidence of the broadening of the taxonomic point of view. Professor Buchanan notes that there has not been any international agreement as to what stage of a bacterium should be designated the mature, adult or perfect stage and that hence there are uncertainties in classification. In fact, all the stages of bacterial development are not yet known, although the records of variability are becoming increasingly extensive. Only a relatively small part of the existing knowledge of bacterial variation has been incorporated in this edition of the manual. It is satisfactory, however, to read that this deficiency is recognized and that "in future editions it is hoped that this lack may be corrected, and descriptions of variants or stages included."

The latest bacteriologic Code is the outcome of the deliberations of the International Society of Microbiology and the International Botanical Congress in 1930. As it is now recognized that the bacteria have characteristics which differentiate them in several respects from both plants and animals, the International Society of Microbiology voted to follow the rules of nomenclature agreed on by international congresses of botany and zoology only "in so far as they may be applicable and appropriate."

According to the count made by the reviewer, this edition of the manual contains descriptions of 1,179 species of organisms (bacteria, actinomycetes and spirochetes), listed in 114 genera. Two new genera have been recognized, namely, the genus *Brucella*, for the organisms of infectious abortion, undulant fever and Malta fever, and the genus *Listerella*, containing the single species *monocytogenes*, the cause of infectious mononucleosis in rabbits. The placing of the *abortus-melitensis* organisms in a separate genus, appropriately named, is an improvement on their former unnatural assignment to the genus *Alcaligenes*, in which they were grouped with the unrelated species *faecalis alcaligenes*. The genus *Pfeifferella*, containing the bacillus of glands has been combined with the genus *Actinobacillus*, including the bacterium of *Lignières* and Spitz. This seems to be an unnatural grouping, from the point of view of pathology. According to the preface, about

50 new species have been included, while several organisms have been omitted as distinct species and their names recognized as synonyms for other species.

The instability of nomenclature is exemplified by these shifting of genera and species and by other editorial as well as taxonomic changes. With the aid of Professor van Eseltine, much has been done to bring "the spelling and endings of scientific names into harmony with the latest international rules" and with Latin grammar. Unfortunately, new names are thus created, and others are to be anticipated. There is still a variation in the use of diphthongs. The genus *Hemophilus* is thus spelled with phonetic simplicity, but *Streptococcus anhaemolyticus* retains the ancient orthographic stem.

Professor Breed's index is of immense assistance to both the experts in the Bergey neologisms and those who adhere to the old familiar terminology in finding the locations of descriptions of bacterial species. The index is an almost complete synonymy.

Opinion on the validity of the classifications and groupings used in this manual will vary according to the points of view of bacteriologists. The distinctions of species among the streptococci are doubtful. *Bacterium tularensis* appears to be related more nearly to the organisms of the *abortus-melitensis* group than to the *Pasteurella* group, to which it is assigned in this book. Undoubtedly, the membership of many groups might be the subject of prolonged debate, with inconclusive results.

Several omissions are notable. The book does not contain descriptions of the spirillum or spirochete of rat-bite fever, the bacillary forms known as *Bartonella* and the bacteria-like organisms now called *Rickettsia*.

In these days of increasing and confusing knowledge of variation, dissociation and antigenic composition, it is probable that most bacteriologists will agree that the bacteria are unclassifiable. On the other hand, they recognize the need of some systematic framework on which to hang, if only temporarily, the shreds of information which are sorted from the sheets of the publications and reports of research. This manual has satisfied that need and has served as an accessible storehouse of older knowledge. The danger exists that the manual, by making a convenient nomenclature available, will tend to fix false notions through everyday usage of names which appear to mean more than they can mean. The uncritical uses of the manual will be exposed to this danger. The editors themselves, however, expressing a liberal point of view and acknowledging that the manual has no "legal" authority, have by their repeated rearrangement of species and genera and their frequent rechristenings of the bacteria, given notice to the reader that each edition has somewhat the characteristics of a progress-report.

The writings of bacteriologists both in this country and abroad indicate that the manual is being used with increasing frequency. This edition was awaited with keen anticipation by many students, and its appearance, with the revised descriptions, is welcomed as a most valuable source of information and an instrument of great service to the science of bacteriology.

The use of a better grade of slightly glazed paper has made the type more legible, improved the appearance of the book and made it more substantial than any of the previous editions.

**Recent Advances in Pathology.** By Geoffrey Hadfield, M.D., F.R.C.P. (Lond.), Professor of Pathology in the University of Bristol, Examiner in Pathology, Late Professor of Pathology, in the University of London, and Lawrence P. Garrod, M.A., M.D., B.Ch. (Camb.), M.R.C.P. (Lond.), Bacteriologist and Lecturer in Bacteriology, Late Demonstrator of Pathology, St. Bartholomew's Hospital. Second edition. Price, \$4. Pp. 457, with 69 illustrations. Philadelphia: P. Blakiston's Son & Co., 1934.

The first edition of this book, which appeared two years ago, received favorable mention in the ARCHIVES (14:589 [Oct.] 1932). The new edition has been revised and enlarged. Among the minor additions may be mentioned statements and discussions concerning thorotrust, monocytic leukemia, cancer caused by radium,

filtrable tumors, resistance to cancer, silicosis, the frequency of cancer of the lung and the results of newer observations on the suprarenal gland in Addison's disease. There is a new section on the pituitary gland, in which the recent advances in its study are presented. The chapter on the liver has been extended by a new section on the recent work on the formation of gallstones. Peptic ulcer and the relations of gastro-intestinal disorders to anemia receive revised and new consideration. The structural changes in the deficiency diseases are well described in the new edition. The chapters on Bright's disease have been rearranged and expanded. It is not practicable to review the seventeen chapters one by one in detail. The book may be recommended as giving convenient, reliable, helpful summaries of recent investigations of the reticulo-endothelial system; tissue culture; cancer; the structural changes in deficiency diseases and important problems connected with the cardiovascular, respiratory, digestive, urinary and nervous systems and the ductless glands.

**Industrial Toxicology.** By Alice Hamilton, M.D. Harper's Medical Monographs. Fabrikoid. Price, \$3. Pp. 352. New York: Harper & Brothers, 1934.

This book aims "to present what is most needed in a short review of modern industrial toxicology rather than a logically planned textbook." The introduction deals with the general methods of preventing industrial poisoning, particularly as concerns the duties and responsibilities of the physician. That the worker in chemical industry shall benefit as much as possible from what is known about industrial poisoning, its prevention and treatment is the basic purpose of the author. There are ten chapters treating of the following: alkalis; acids; chromium; lead; arsenic; mercury; metal-fume fever; other metals (copper, phosphorus, zinc, etc.); asphyxiants (carbon monoxide and dioxide, hydrogen sulphide and cyanide); benzene and other derivatives of coal tar; petroleum and derivatives; turpentine; carbon disulphide; tobacco; mineral oils; occupational cancer, and radio-active substances. There is an extensive bibliography (to January 1933), which will be of great help. The book is well written. Within a small compass it contains a great deal of reliable information about industrial poisoning. It reveals that industrial chemical methods change, that new chemicals come into use, that old chemicals are used in new ways and that dangers of industrial poisoning continue. The book will be of value to all who are concerned with such problems; it supplements nicely the larger book by the same author ("Industrial Poisons in the United States," New York: The Macmillan Company, 1925).

## Books Received

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LA SÉRORÉACTION BLENNORRAGIQUE. Bertrand Reme. Price, 30 francs. Pp. 179. Paris: Masson & Cie, 1934.

Here is a full account of the history, the technic, the evolution, the specificness and the practical value in diagnosis, prognosis and prophylaxis of complement fixation in gonococcic infection. The bibliography is as complete as possible. Technical matters are discussed in detail, and the importance of standardized, stable antigen is emphasized.

DYSENTERY IN DENMARK: A CONTRIBUTION TO THE BACTERIOLOGY AND EPIDEMIOLOGY OF INFECTION WITH SONNE AND FLEXNER BACILLI. Communications de l'Institut Sérothérapique de l'Etat Danois, tome 24, 1934. Knud Bojlén. Pp. 231. Copenhagen: Bianco Lunos, 1934.

PARASITISM AND DISEASE. Theobald Smith, Director Emeritus of the Department of Animal Pathology, Rockefeller Institute for Medical Research. Published by the Louis Clark Vanuxem Foundation. Price, \$2. Pp. 196. Princeton: Princeton University Press, 1934.

INDUSTRIAL TOXICOLOGY. Harper's Medical Monographs. Alice Hamilton, M.D. Price, \$3. Pp. 352. New York: Harper & Brothers, 1934.

DISEASES PECULIAR TO CIVILIZED MAN: CLINICAL MANAGEMENT AND SURGICAL TREATMENT. George Crile, M.D. Edited by Amy Rowland. Price, \$5. Pp. 427, with 41 illustrations. New York: The Macmillan Company, 1934.

# ARCHIVES OF PATHOLOGY

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## EXPERIMENTAL STUDIES ON HUMAN AND PRIMATE SPECIES OF STRONGYLOIDES

### III. THE FECUNDITY OF STRONGYLOIDES FEMALES OF THE PARASITIC GENERATION

ERNEST CARROLL FAUST, PH.D.

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CORINE ADAMS, M.S.

AND

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NEW ORLEANS

In our earlier studies on experimental Strongyloides infections in man, in dogs and in several species of monkeys, we were particularly concerned (1) with the type of strain as it developed in culture and as it was subinoculated successively from one host to the next,<sup>1</sup> and (2) with the consecutive stages in the development of the parasitic generation, including both females and males.<sup>2</sup> It soon became evident that the strains utilized manifested marked variability and instability in type. There appeared to be some evidence of correlation on the one hand between indirect strains and fertilized eggs of the parasitic worms, and, on the other, between direct strains and unfertilized eggs. While this correlation was not a matter of direct experimental proof, evidence favoring this view was submitted for consideration.<sup>2</sup>

The problem was complicated by other factors. In dogs of the same size, weight and age, fed the same rations, inocula of filariform larvae from the same cultures in similar amounts, introduced by the same route, resulted in infections which produced different daily and average yields of rhabditiform larvae in the evacuated stools. Furthermore, autopsy on the experimental hosts revealed a difference in the egg-producing capacity of the parasitic females. After several months of infection some worms were maintaining a high productivity; others were producing a small number of eggs, and still others were apparently completely unproductive.

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Contribution from the Parasitology Laboratory, Department of Tropical Medicine, Tulane University of Louisiana.

This investigation was supported by a grant from the Committee on Scientific Research of the American Medical Association.

1. Faust, E. C., and Kagy, E. S.: Am. J. Trop. Med. **13**:47, 1932.

2. Faust, E. C.: Am. J. Hyg. **18**:114, 1933.

In an attempt to discover the basis for these differences in the fecundity of parasitic females in human and nonhuman strains of *Strongyloides*, series of controlled experimental infections were undertaken.<sup>3</sup>

#### MATERIAL AND TECHNIC

The data presented in this communication were obtained from a study of seventeen pups (from 3 to 6 months old at the time of inoculation) inoculated with human strains of *Strongyloides stercoralis*, and one 5 year old *Pithecius rhesus*, inoculated with a chimpanzee strain, morphologically and physiologically identical with human strains (indirect type) of *S. stercoralis*. In the canine series, previous to inoculation the animals were devocalized, treated with tetrachlorethylene for the eradication of hookworms and tested by intensive fecal examination and culture to make sure, so far as antemortem tests could indicate, that they did not harbor *Strongyloides*. Seven animals (nos. 42 [second inoculation], 43, 45, 47, 48 [twice], 56 and 59) were inoculated by the oral route; eight (nos. 41, 42 [first inoculation], 57, 60, 61, 62, 63 and 64), by the abdominal cutaneous route, and three (nos. 50, 54 and 58), by the intracecal route. Except for nos. 56, 57, 58 and 59, in which second generation (indirect type) filariform larvae were used, all the animals were inoculated with first generation (direct type) filariform larvae ( $f_1$  or  $F_1$ ). In the case of dog 47 the inoculation was obtained after passage through dog 45, and in dog 59, larvae obtained from the feces of dog 48 were used as the inoculum. All the remaining dogs were inoculated with larvae obtained directly from cultures of human stools. Five human strains were utilized (strain M, six animals; strain O, six animals; strain P, two animals; strain Q, four animals, and strain R, one animal; two duplicate inoculations were made). A fecal examination of each animal was made daily for the period from two weeks before inoculation until autopsy.

As soon as the animal showed evidences of harboring the organism (or, in certain exceptions, somewhat later) the fresh daily fecal evacuation was weighed; a 5 Gm. portion was carefully screened through wire gauze and concentrated, and the total number of larvae (or eggs) in the sample were counted. In this way the total daily yield of the progeny was calculated, with an estimated error usually not over 10 per cent. Although this count of the total number of larvae or eggs present in the 5 Gm. sample was exceedingly tedious, it frequently demonstrated their presence when three ordinary fecal samplings were negative.

The greatest difficulty arose in detecting dead and degenerate larvae in the stool. It was found necessary to dilute the concentrate sufficiently so that fragments of larvae were not obscured by débris. Because of the percentage of dead and degenerating larvae in freshly passed feces (of man and dogs), ranging at various times from 5 to 90 per cent, it was found that Sandground's technic<sup>4</sup> for culturing the progeny was entirely unreliable in providing a basis for estimating the total production of the parasitic females. On a few occasions the feces were found contaminated with *Rhabditis* larvae from the perianal hairs, even though the animals and their cages were regularly scrubbed with a strong solution of cresol. When such contaminations occurred the counts had to be discontinued for two or three days.

3. Faust, E. C.; Wells, J. W.; Adams, C., and Beach, T. D.: Proc. Soc. Exper. Biol. & Med. **31**:1041, 1934.

4. Sandground, J. H.: Am. J. Hyg. **6**:337, 1926; **8**:507, 1928.

All the animals in the series were killed. The following technic was carried out at autopsy: On opening the abdominal cavity the gastro-intestinal tract was tied off above the cardiac sphincter and at the anus and removed as a complete viscus. Similarly, above the diaphragm, the lungs, bronchi, trachea, glottis and esophagus were removed en masse. The stomach and intestines were opened from above, and the work was carried on distalward. The contents of the lumen of each level were first washed out into physiologic solution of sodium chloride; following this, scrapings were made from the mucosal surface successively deeper and deeper down to the muscular coats. (Experience has shown that the worms rarely penetrated below the muscularis mucosae). Approximate estimates of the numbers of eggs and larvae and exact counts of adult worms were made for successive levels of the entire lumen and of the mucous and submucous coats of the entire stomach and intestines, so that the total number of parasitic females was believed to have been discovered. The trachea and bronchi were opened from above downward into the bronchioles. Washings and scrapings were made along the entire respiratory tree and examined microscopically. Scrapings were then made from representative sections of the lungs following which the lungs were chopped up, left in physiologic solution of sodium chloride in the electric ice-box overnight, squeezed and strained out the next morning, and the semiliquid yield centrifugated. The centrifugate was then completely examined under the microscope for adult worms and larvae. The esophagus was washed and scraped in a similar manner. When females were recovered, a careful study was made of their condition—whether active, semiactive or dead, whether mature or immature—and more particularly efforts were made to discover indications of continued or reduced productivity in mature females, as determined by eggs or larvae in utero and in the immediately adjacent tissues of the host.

#### PRESENTATION OF DATA

A. HUMAN INFECTIONS IN DOGS.—The canine series was divided into three sub-series: (1) those which were negative for parasitic females at autopsy; (2) those with relatively light infections (200 or less females per animal), and (3) those with relatively heavy infections (more than 200 females per animal).

1. *Animals Showing No Evidence of Organisms at Autopsy*.—Dog. 54: The animal was inoculated intracecally with 250 active  $F_1$  larvae, strain O (six day culture). Organisms were found only once, one hundred and forty-three days after inoculation. The animal was killed on the two hundred and third day. No Strongyloides, adults or larvae, were found on thorough examination of the organs, except one dead filariform larva from the right lung (believed to have been a contamination, but possibly the progeny of a female undetected in the postmortem search).

Dog 42: The animal was inoculated on the skin of the abdomen with 5,000 active  $F_1$  larvae, strain M (three day culture). The prepatent period was sixteen days. The animal showed Strongyloides five times during the next fifty-five days. It was reinoculated on the fifty-fifth day with 500  $F_1$  larvae (strain O, one day culture) in aqueous suspension introduced into the buccal cavity. The prepatent period of the second infection was twenty-four days, as indicated by the large number of larvae which appeared for the first time at the end of that period. Larvae were again found twice within the next six months. The animal was killed nine months after the original inoculation. The postmortem search for Strongyloides gave negative results.

2. *Animals with Relatively Light Infection.*—Dog 45: The animal was inoculated orally with 250  $f_1$  larvae (five day culture, strain M). The prepatent period was sixteen days. The animal showed a slight evidence of infection on five isolated days within the next one hundred and thirty days. It was killed on the one hundred and thirty-first day. Forty-four nonfecund parasitic females were found as follows: duodenum, 10; jejunum, 24; ileum, 3; concentrate of intestinal washings, 7. The lungs were normal. An example of the females obtained from the intestine is shown in figure 1. Uterine cords had developed but had not yet formed hollow tubules; no eggs were found. All the body characters were indicative of a young adolescent parasitic female. The case is interpreted as one in which the original infection had died out and a more recent

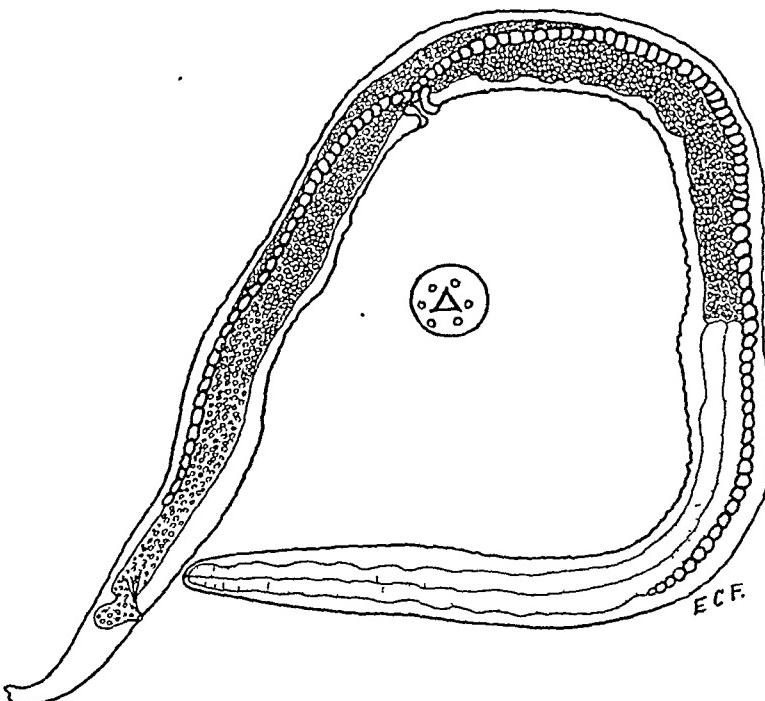


Fig. 1.—Adolescent female *S. stercoralis* removed from deep scrapings of the duodenum of dog 45, two hours after the host was killed on the one hundred and thirty-first day after inoculation. The midintestine and rectum of the parasites are filled with granular material. The vulva is readily distinguished on the ventral (incurved) side, just behind the middle of the worm. The two horns of the uterus, with their oviduct-ovarian continuations, are still solid cords and have not yet become hollow tubules. In the center of the illustration is a head-on diagrammatic representation of the buccal opening, surrounded by six oral papillae. Drawing by means of camera lucida;  $\times 200$ .

(hyperinfective?) group of females had not yet reached maturity. All the worms were apparently in the same stage of development.

Dog 43: This dog was inoculated orally with 1,000  $f_1$  larvae (strain O, one day culture). The prepatent period was eighteen days. The results of examination were positive on six isolated occasions during eighteen days. The animal was killed on the one hundred and sixty-fifth day. Twenty-seven females were found as follows: duodenum, 22; jejunum, 4; concentrate of intestinal wash-

ings, 1. Only 3 worms contained eggs, 2 each. No eggs or larvae were found in the concentrates of the lumens or in the scrapings from tissue. The lungs were normal. The yield of females constituted only 2.7 per cent of the number of filariform larvae in the inoculum.

Dog 64: The animal was inoculated by the abdominal cutaneous route with 1,500  $f_1$  larvae from a one day culture (strain Q). The prepatent period was eleven days. The calculated total numbers of larvae in evacuated feces for the next thirty-one days were as follows: 16; 57; 1,224; 80; 24; —; 0; 0; 78; 0; 216; 0; 0; —; 14; 0; 0; 0; 0; 10; —; 0; 0; 0; —; 0; 0; 0; 0; 0; 0.<sup>5</sup> The animal was killed on the forty-second day after inoculation. One hundred and sixty-six female worms were recovered as follows: duodenum, 73; jejunum, 77; ileum, 3; concentrate of intestinal washings, 13. Less than 10 per cent of the worms contained eggs (from 1 to 3 each). Most of the worms were encapsulated. No larvae were seen in the tissues or concentrates. The lungs were normal. The yield of parasitic females recovered was 11 per cent of the larvae used in the inoculum.

Dog 48: The animal was inoculated orally with 1,500  $F_1$  larvae (one day culture, strain M). The prepatent period was thirteen days. Larvae were found again in stools only on two succeeding days after the period of incubation. On the nineteenth day the animal was reinoculated by mouth with 500  $f_1$  (four day culture, strain O). The prepatent period of the second strain was fourteen days. For two weeks thereafter large numbers of larvae were recovered from the stools. After three months 5 Gm. samples of feces were consistently negative. The animal was killed on the one hundred and fortieth day after the original inoculation. One hundred and forty-four female worms were recovered from the following levels of the intestinal tract: duodenum, 69; jejunum, 71; ileum, 1; concentrate of intestinal washings, 3; lungs, 1 female and 1 preadolescent male worm. Except for a single specimen which had a few eggs in utero, all the females were active but were apparently nonfecund. No eggs or larvae were found in the tissues.

Dog 56: It was inoculated orally with 1,000  $f_2$  larvae (four day culture, strain P). The prepatent period was thirteen days. Larvae were recovered again only on four isolated occasions during the next thirty-six days. The animal was killed one hundred and ninety days after inoculation. Only 3 parasitic female worms were recovered as follows: duodenum, 2; ileum, 1. They were all encapsulated, and none of them contained eggs. No eggs or larvae were found in the surrounding tissues. However, two viable filariform larvae of *Strongyloides* were recovered from lung concentrates.

Dog 57: The animal was inoculated by the abdominal cutaneous route with 500  $f_2$  larvae (three day culture, strain O). The feces never showed the presence of the organism. The animal was killed six and a half months after inoculation. Six nonfecund encapsulated female worms were recovered as follows: duodenum, 2 (each with 1 degenerating egg in utero); jejunum, 2; concentrate of intestinal washings, 2. The lungs were normal.

Dog. 47: This dog was inoculated orally with 200  $F_2$  larvae (one day culture, strain M). The prepatent period was fifteen days. The total numbers of calcu-

5. A dash indicates that no stools were passed.

3. *Animals with Relatively Heavy Infection*.—Dog 61: The animal was inoculated by the abdominal cutaneous route with 1,500 f<sub>1</sub> larvae (four day culture, strain Q). The prepatent period was eleven days. The number of larvae calculated to have been passed in evacuated feces for seventy-six consecutive days was as follows: 253; 42,728; 202; 4,715; 6,094; 3,365; 1,265; 3,078; 32,929; 1,823; 25,519; —; 5,531; —; 12,070; 62,496; 3,504; 14,033; 1,760; 6,282; 12,642; 2,230; 33,886; 5,876; 122,262; 102,500; 78,019; 108,720; 61,700; 95,900; 63,900; 80,700; 97,950; 114,150; 33,750; 4,500; 44,800; 6,160; 19,650; 53,550; 21,450; 38,850; 25,600; 20,400; 21,450; 18,150; 5,000; 15,150; 10,950; 5,000; 3,300; 2,800; 4,200; 1,700; 14,250; 6,300; 2,800; 1,500; 2,860; 3,500; 1,500; 1,700; 3,500; 2,400; 600; 2,550; 1,100; 0; 0; 0; 600; 800; 150; 300.<sup>5</sup> The animal was killed on the eighty-fifth day after inoculation. Six hundred and eight female worms were recovered as follows: duodenum, 91; jejunum, 392; ileum, 63; concentrate of intestinal washings, 62. Only 1 egg and no larvae were found in the abdominal viscera examined; the egg was lodged in the tissues; all the females were nonfecund and encapsulated. The thoracic viscera were normal.

Dog 59: This dog was inoculated orally with 2,000 f<sub>2</sub> larvae (six day culture of strain O, through dog 48). The prepatent period was twelve days. The numbers of larvae calculated to have been passed in the evacuated stool for thirty-seven consecutive days after the prepatent period were as follows: 41; 3,758; 6,050; 9,786; 396; 1,152; 8,597; 8,864; 1,061; 8,659; 1,031; 16,541; 2,136; 4,598; 2,494; 19,734; 11,380; 12,833; —; 26,296; 2,659; 2,672; 5,930; 12,642; —; 7,912; 2,482; 10,512; 8,332; 2,150; 6,415; 644; 366; 3,481; 770; 785; 2,479.<sup>5</sup> The animal was killed on the forty-ninth day after inoculation. Seven hundred and eight female worms were recovered as follows: duodenum, 109; jejunum, 293; ileum, 171; concentrate of intestinal washings, 135. Thirty-seven rh<sub>1</sub> larvae were found in the pyloric wall of the stomach, but no adult worms were discovered there. The reaction of gastric juices was weakly basic. The number of larvae seen in the intestine proper was somewhat smaller than the yield of females. The females recovered from scrapings of living tissue in the duodenum and upper jejunum were deeply embedded in the mucosa. Some were surrounded by epithelial envelopes. Only a few eggs were found in a small proportion of the females; most females were apparently postproductive. The thoracic viscera were normal.

Dog 41: The animal was inoculated by the abdominal cutaneous route with 2,000  $f_1$  larvae (four day culture, strain M). The prepatent period was fourteen days. The feces were strongly positive for four months following the first recovery of larvae. The calculated counts of larvae, based on weighed samples for eight days at the end of this period, were as follows: 7,779; 4,613; 12,305; 2,073; 5,904; 5,409; 3,155; 2,295. The animal was killed five months after inoculation. Eight hundred and forty-four female worms were recovered from the intestine as follows: duodenum, 318; jejunum, 374; ileum, 118; concentrate of intestinal washings, 34. The thoracic viscera were normal. The eggs in utero and both eggs and larvae in the tissues were numerous. There was no evidence of

greatly diminished fecundity in mature worms, although about 25 per cent were immature or had apparently just commenced to lay eggs.

Dog 60: It was inoculated by the abdominal cutaneous route with 750  $f_1$  larvae (one day culture, strain Q). The prepatent period was five days. The calculated total numbers of larvae for thirty-seven successive days following patency were as follows: 6; 51; 0; 39; 14; 0; 122; 0; 90; 7; 0; 41; 17; 0; 0; 10; 108; 0; 5; 0; 8; 0; 0; —; 0; 44; 48; 0; 241; 1; 125; 67; 32; 14; 65; 34; 88.<sup>5</sup> On the twenty-eighth day of this count one active parasitic female, containing two unembryonated eggs in utero, was recovered from the fecal concentrate. The dog was killed on the forty-first day after inoculation. Eight hundred and twenty-one female worms were recovered from the intestine as follows: duodenum, 186; jejunum, 500; ileum, 79; concentrate of intestinal washings, 56. One filariform *Strongyloides* larva was recovered from the left lung. The great majority of the females were postproductive, and some were surrounded by epithelial envelops.

Dog 50: The animal was inoculated intracecally with 100  $f_1$  larvae (strain M, passed through dog 41). The prepatent period was eleven days. Larvae were found occasionally in the feces during the next fifty days. The calculated numbers of larvae for the next six days (fifty-first to fifty-seventh day) were as follows: 624; 167; 20; 574; 638; 300. The animal was killed on the one hundred and twentieth day after inoculation. Two hundred and fourteen parasitic females were recovered as follows: duodenum, 71; jejunum, 104; ileum, 16; concentrate of intestinal washings, 25. The worms usually contained a few (rarely more than three) formed eggs in utero. Unformed or infertile eggs were frequently found in tissues immediately surrounding the females, along with smaller numbers of embryonating eggs. A few recovered females were in the adolescent stage. No worms were found in the lungs, but a few embryonating eggs were obtained from concentrates of each lung, apparently indicating the presence in each organ of at least one fertile female which had been overlooked. The data definitely indicate a hyperinfection, with a yield of adult females amounting to more than twice the number of  $f_1$  larvae in the original inoculum.

Dog 62: This dog was inoculated by the abdominal cutaneous route with 2,000  $f_1$  larvae (strain Q). The prepatent period was thirteen days. The number of larvae on the day following the first recovery of larvae in stools was calculated to be 525. The animal was killed on the next day. Two hundred and forty-eight unencapsulated female worms were recorded from the intestine as follows: pylorus, 1; duodenum, 134; jejunum, 90; ileum, 4; concentrate of intestinal washings, 19. Eggs and motile  $rh_1$  larvae were numerous at various levels of the bowel. The thoracic viscera were normal.

Dog 63: The animal was inoculated by the abdominal cutaneous route with 14,000  $f_1$  larvae (one day culture, strain R). The prepatent period was fourteen days. The daily computations of the total number of larvae in the feces for twenty-six days from the beginning of patency were as follows: 220; 0; 35; 54; 54; 0; 42; 52; 22; 0; 0; 0; 1; 934; 19; 0; 0; 0; 45; 0; 0; 0; 0; 0. The animal was killed on the day after the last count was made (forty-one days after inoculation). Four hundred and sixty-two female worms were recovered from the intestine as follows: duodenum, 112; jejunum, 303; ileum, 29; concentrate of intestinal washings, 18. Most of the worms contained only a few eggs in utero; some worms were already definitely postfunctional, as indicated by the complete absence of egg-forming material in the ovaries and by the transparent, glossy consistency of all their tissues and organs; the worms, however, were still alive. All these worms

were enveloped in an epithelial capsule. Tissues immediately surrounding the worms contained only a few eggs and larvae.

Dog 58: It was inoculated intracecally with 2,000  $f_2$  larvae (five day culture, strain P). The prepatent period was twelve days. The stools were positive daily for twelve days, later they were irregularly positive as shown by the routine smear method. Computations of the number of larvae, based on daily counts for seven days, beginning seventy days from the date of patency were as follows: 178; 102; 136; 138; 112; 34; 0. The animal was killed on the eighty-ninth day after inoculation. Two hundred and ten female worms were recovered from the intestine as follows: duodenum, 58; jejunum, 117; ileum, 24; concentrate of intestinal washings, 11. Nearly all these worms were enveloped in an epithelial capsule (fig. 2). Few eggs were present in utero or in the tissues of the host immediately surrounding the worms. Fewer larvae were found in the tissues. Only four worms were definitely found to be still productive. Most worms were postproductive, with the integument greatly contracted and the protoplasm transparent, although such worms became very active when removed to physiologic solution of sodium chloride. A small portion of the yield was diagnosed on morphologic characters as preadolescent or adolescent. The worms recovered were roughly grouped as follows: (1) preadolescent and adolescent females, 15 per cent; (2) productive females, 2 per cent, and (3) postproductive females, 83 per cent. The pulmonary tissues gave the following yield: left lung, 1 preadolescent female; right lung, 7 postfilariform larvae (sex undetermined).

B. STRONGYLOIDES IN THE MONKEY.—This work was carried out on a five year old Pithecius rhesus, which had been under laboratory observation for four years and had never showed evidence of harboring Strongyloides during this period. The animal was inoculated orally with 24,000  $f_2$  larvae of a chimpanzee strain, which had been studied for four months previously in the original host as well as in a rhesus monkey and in a human volunteer. The strain was known to be a pure indirect type. The animal showed evidence of harboring the organism after a seventeen day period of incubation. The number of eggs and larvae in the stools gradually increased for the next month. After four months the calculated number per twenty-four hour sample amounted to about 1,000,000. During this period the monkey had a leukopenia (25 per cent) with marked eosinophilia (18.5 per cent). Daily examinations were continued for the next five months, but quantitative studies were not resumed until the end of that period, when counts indicated a greatly reduced first generation progeny, as demonstrated by the following total daily calculated output over a period of twenty-nine days: 3,480; 21,608; 10,250; 38,000; 13,150; 83,600; 38,350; 21,875; 12,800; 12,600; 26,800; 9,500; 36,400; 5,704; 22,800; 22,050; 36,800; 33,600; 38,800; 33,300; 22,000; 28,600; 33,920; 32,400; 32,200; 32,900; 23,700; 40,808, and 25,500. After a three week intermission another five week count showed about the same productive level, with comparable daily variations. Fourteen months after inoculation the average daily count became noticeably lower, as follows: 12,600; 12,000; 10,000; 1,350; 22,000; 18,000; 11,200; 20,700; 27,500, and 10,800.

The host was killed fourteen and a half months after inoculation. At autopsy 523 unencapsulated parasitic females were recovered from the following levels in the intestinal wall: pylorus, 1; duodenum, 376; jejunum, 110; ileum, 7; cecum, 1; concentrate of intestinal washings, 28. Large numbers of eggs were present in the lumen and in the immediate vicinity of the worms in the tissues. While eggs in utero indicated that practically all the parasitic females were still productive, the number of such eggs per female was small, usually only 2 or 3 and never over 10. No worms, eggs or larvae were found in the thoracic viscera.

The figures for the infection in the monkey show a maximum daily computed number of 1,000,000 progeny one hundred and thirty-eight days after inoculation; a marked reduction to an average level of 25,000 eggs, with considerable daily fluctuations, after ten months; a continued production of eggs at approximately this level for about three months, and a subsequent drop in the level to an average of 13,250. On the basis of parallel examinations of another rhesus monkey inoculated with this chimpanzee strain, it may be concluded that the fertility of the parasitic females would have ceased in the following two or three months, after which there would have been no further evidence from the feces of the presence of worms in the intestinal wall.

At the time the average daily output of first generation progeny was 25,000 the daily output per worm was calculated to be approximately 50; later it dropped to about 27. These figures, which are much greater than the maximum number of eggs in utero in the parasitic females at autopsy, indicate either that the production of eggs and parturition occurred considerably more frequently than once daily, or that the worms were much less productive at autopsy than they were during the periods when the counts were made. This question will be considered later.

#### GENERAL CONSIDERATIONS

A casual examination of the data which have been presented indicates that in *Strongyloides* infections in the experimental host processes are at work which involve both the parasite and the host. In the first place, our studies, like those of previous investigators, demonstrate that infection may be acquired either cutaneously or orally. In the latter case it may be temporarily assumed that the filariform larvae invade the venous blood stream via the buccal, pharyngeal and esophageal mucosa. Experimental proof of this assumption has been obtained by us and will be published elsewhere. In experimental animals both methods of inoculation are equally successful, although it is likely that in nature the cutaneous route is by far the more common. Furthermore, we have demonstrated that infection may be readily established by introducing larvae in the infective stage into the lumen of the large bowel. In this demonstration of how internal infection (hyperinfection) may occur, the greatest precautions were exercised to prevent any larvae returning through the anus from penetrating through the perianal skin, by keeping this area thoroughly saturated with a strong solution of iodine for several hours after the inoculum had been introduced into the cecum. Infections may therefore be established by three portals of entry: (1) the skin, (2) the pregastric mucosa and (3) the intracecal route.

One of us<sup>2</sup> has previously shown the successive stages in the development of the adult female and male parasitic *Strongyloides*, including

the filariform and postfilariform larvae, the preadolescent, adolescent and mature worms, and, later, the postfunctional and degenerate females. Evidence favors the view that females, if fertilized by the "ephemeral" males, are apparently impregnated in the adolescent stage, either in the bronchioles of the lungs or before they enter the intestinal mucosa. While the intestinal wall is the common habitat of adult worms, it has been shown that the bronchioles of the lungs are a suitable site for their complete development and that this focus may be a source of subsequent hyperinfection. Females may become mature and begin to produce eggs in the lungs as early as the third day after inoculation; in the intestine they may produce eggs as late as the twenty-seventh day.

Females may begin to lay eggs before actually penetrating into the intestinal mucosa, but this is not common. In a previous series of experiments some of the canine hosts were killed between the fifth and the ninth day after inoculation. At this stage of the infection the animal frequently had a prodromal diarrhea. The duodenum and jejunum, when opened, were usually hyperemic and hemorrhagic. Surface scrapings of the mucosa ordinarily yielded a film of mucus in which there were many adolescent and maturing females and, less commonly, adolescent and mature males. Free eggs and rhabditiform larvae were not numerous in the film (not over 2 or 3 per female worm). A few developing eggs were always present in each uterine arm of young mature worms. This condition preceded by one or two days the appearance of rhabditiform larvae (less frequently of eggs) in the fecal dejecta. From that time on for several days it was found that the females penetrated more deeply into the mucosa, so that, when the animal was killed, successively deeper scrapings of this layer were required to obtain a maximum yield of adult females and larvae.

When the females had become well established in the mucosa of the intestinal wall, parturition rapidly rose to a maximum; then, after a period of days, weeks or months, it descended to a level which was much lower, but still greatly in excess of the total number of uterine eggs in the female worms. Later this production of eggs was gradually reduced to a lower and lower level, and finally it ceased. Although individual counts of progeny produced by the mother worms seldom coincided in any two canine hosts, under conditions as comparable as it was possible to obtain, the sequence of events was essentially the same. In some animals the period of egg production was apparently limited to weeks; in others, to months. Since such discrepancies existed in animals inoculated with homologous as well as with heterologous strains, it is suggested that the determining factor is probably a reaction of the tissue of the host to the worm and is not intrinsic in the worm itself. In the infection of the monkey, after declining from a high maximum, the

moderate level of egg production was maintained over a period of many months, but later it showed a definite evidence of diminution.

The canine series in this study has been presented more or less in reverse order to the fecundity of the worms (see Presentation of Data, A). Dogs 54 and 42 did not show evidences of *Strongyloides* at autopsy; dogs 45, 56, 57 and 61 had only nonfecund females; dogs 43, 64, 48, 47, 59, 60, 50, 63 and 58 yielded females which were mostly nonfecund, while dogs 41 and 62 harbored worms which showed evidence of continued fecundity. The productivity of these females was not correlated with the strain of the organism, nor was it always related to the length of the infection within the host, although the general tendency was a definite decline in the production of eggs after a period of months. Taken as a whole, the productive period of human *Strongyloides* in dogs is relatively short, a matter of months, as contrasted with a period presumably of years in man, so that the complete life cycle is considerably abbreviated. This has both its advantage and its disadvantage: it is of advantage in demonstrating the complete picture of development and egg production in a relatively short time, but of disadvantage in making a direct comparison with human infections more difficult.

The percentage of female worms recovered at autopsy, as compared with the number of filariform larvae present in the inoculum, varied enormously. When dogs 54 and 42, from which no worms were recovered, are omitted, the yield ranged from 0.3 per cent (dog 56) to considerably over 100 per cent (dog 60, 109.5 per cent; dog 50, 214 per cent). The majority, however, varied from 10 to 40 per cent. Since experience has shown that a large number of filariform larvae are filtered out in the skin when the inoculum is applied to this portal of entry,<sup>6</sup> a yield of parasitic females amounting to between 10 and 20 per cent of the filariform larvae used in cutaneous inoculations is considered to be satisfactory. This series of experiments indicates that oral or intracecal administration of the inoculum gives results comparable to those obtained by cutaneous inoculation. In each series there are cases with more than a 20 per cent yield (cutaneous route, 3, 42.4 per cent of animals employed; oral route, 1, 16.6 per cent, and intracecal route, 1, 50 per cent). Since the parasitic females recovered at autopsy represent the minimum and not the maximum number of females which actually matured, and since allowance must also be made for parasitic males, any excess of worms above 20 per cent of the larvae in the inoculum requires analysis. Certainly yields above 100 per cent demand a careful scrutiny. It is not likely that any considerable error (10 per cent or more) resulted from underestimating the number of larvae in

6. Stumberg, J. E.: Am. J. Hyg. 15:186, 1932. Faust.<sup>2</sup>

the inoculum, since two measured samplings of the larvae in each inoculum were carefully counted immediately before the inoculation was undertaken. A portion of such larvae was believed to consist of potential males, and was, therefore, not accounted for in the counts at autopsy made any considerable length of time after the prepatent period.<sup>2</sup> Thus in five animals in the series the actual yield of parasitic females at autopsy is far in excess of expectations. In our opinion this can be accounted for in one way only, namely by internal infection (hyperinfection). Information supporting this view is furnished from two sources. In the case of dogs 50 (intracecal inoculation) and 47 (oral route) on at least one occasion active dwarf filariform larvae ( $f_1$ ) were recovered from the freshly passed stool which still retained the body warmth of the host. In the female worms recovered from dogs 41 and 58 (on which autopsy was done respectively five months and eighty-nine days after inoculation) there was in each case a definite evidence of premature or recently matured female worms (dog 41, 25 per cent of the yield; dog 58, 15 per cent of the yield). In dogs 50 and 47 there was a clue as to the method of internal infection (i. e., dwarf filariform larvae within the bowel in a favorable position to invade the intestinal mucosa). That this method is possible is well illustrated by the positive results of intracecal inoculation of dogs 50 and 58, as well as of other dogs not included in this series. In dogs 41 and 58 there was evidence of recent development of female parasites. While actual proof was not obtained in the case of other animals in which an unexpectedly high yield of parasitic females was obtained at autopsy, it is logical to believe that in the case of those animals, too, the number of organisms was increased by the mechanism of internal infection.

In the section on technic we have referred to the difficulties encountered in obtaining data on the daily productivity of the female worms from examination of the feces. Three fecal smears frequently failed to reveal a single egg or larva. For this reason as well as in order to obtain a quantitative estimate of the daily discharge of eggs in the feces, it was soon discovered that another technic must be employed. Cultures of the material were most unsatisfactory, not only because some of the larvae died in the culture, but more particularly because many larvae were frequently dead when the stools were passed. Even in samplings taken from the upper colon and cecum, at times as many as 80 per cent of the larvae were nonviable. It was, therefore, necessary to make daily counts of weighed samples; these were washed, concentrated and then spread out sufficiently thin on fecal slides so that no worm or fragment of a worm was overlooked in the count. From these counts the total daily discharge of progeny in the feces was reckoned, since the weight of the total fecal discharge was obtained as a routine. We believe, there-

fore, that our calculations are reasonably accurate, although they are minimum rather than total, since it is not unlikely that some larvae had completely disintegrated within the bowel, while there is concrete evidence indicating that in two cases some rhabditiform larvae transformed into the filariform stage within the bowel and may have penetrated into the intestinal mucosa. Nevertheless, we believe that counts of larvae discharged in the feces, no matter how accurate, constitute an unsatisfactory criterion of the daily output of eggs or larvae by the parasitic female. Our conclusions are based on the following grounds:

In the first place, although each dog and the monkey in our series usually had a daily fecal discharge, there were days when no defecation occurred. More commonly the stools were fairly regular and equal in amount, but the numbers of larvae expelled were inconstant. Any variation in larval counts, such as that for dog 59, indicates the irregularity of such discharges of larval progeny. We have been unable to find any intrinsic or extrinsic evidence in the levels of the small intestine, where the majority of the females reside, which explains this irregularity satisfactorily. Only one fact bearing on the situation is known, namely, that the average daily output of larvae (or eggs), as measured either by those counted in the stools or by those present in the lumen of the duodenum and upper jejunum, is far in excess of the number of eggs present in the uteri of fecund females, even at the peak of egg production. By the time the females have become parturient they are usually well embedded in the mucosa, with their vulvae in such a position that eggs are shed into the deeper levels of the mucosa, and not directly into the lumen of the bowel. These eggs may be in the "tadpole" stage of development, but more commonly they contain rhabditiform larvae nearly ready to hatch. Rarely they hatch in utero. Hatching ordinarily takes place soon after the eggs are deposited in the tissues, but some eggs may be discharged into the lumen of the bowel before hatching occurs. The larvae in the tissues may soon work their way out of the wall into the lumen of the bowel or may remain for weeks as active forms, feeding on the cells of the mucosa. Whether in the mucosal layer or in the lumen of the upper small intestine, they grow in size but have never been found to metamorphose into filariform larvae at this level. Farther down the intestine, particularly in the colon and rectum, as well as in the expelled feces, they become appreciably smaller than they were in the duodenum or jejunum. Some of the larvae, which remain in the tissues for weeks, become encased in delicate hyaline capsules; this tends to prolong their stay in the intestinal wall.

It is evident, therefore, that the several conditioning factors which govern the discharge of larvae from the tissues into the lumen of the bowel, the rapidity of movement of the contents down the bowel and

the death and disintegration of the larvae in the lower part of the bowel make it impossible to judge the egg-laying capacity of the females by the daily discharges of larvae in the feces. Careful counts of measured samples of feces over a period of weeks furnish approximate information of productivity, particularly when such counts are conducted at various intervals during the egg-laying period. Because of the difference in egg production at different times during this period, the larvae discharged in the stool furnish no criterion of the number of female worms actually present in the upper part of the intestinal wall. These observations stand in marked contrast to those for hookworms, *Ascaris* and *Trichocephalus*, for which a relatively accurate estimate of the number of female worms present in the bowel may be made by egg counts of measured samples of feces. However, in the case of *Strongyloides*, egg or larval counts of measured samples of feces over a period of several days furnish an approximate idea of the egg-laying capacity of the mother worms at the particular period in the productive cycle.

In view of these observations on egg production in dogs infected with human strains of *S. stercoralis* and in a rhesus monkey infected with a chimpanzee strain of *Strongyloides* morphologically and physiologically resembling human strains of *S. stercoralis*, the question arises as to what conditions are responsible for reduction in fecundity. First of all, as we have indicated previously, the period of egg production of *S. stercoralis* in the dog is relatively short compared with that in the human host. We regard this mechanism in the two hosts as qualitatively similar but differing as to time elements. A month's period in the dog may be comparable to a year's duration of the same strain in man, while in the monkey the productive period apparently lies between these extremes. We have demonstrated that the period of fecundity of parasitic females in homologous human strains varies within wide limits in different dogs. Hence two separate factors apparently operate in determining the actual process, namely (1) the compatibility of the host species and (2) the relationship of the individual host to the parasite. So far as we have been able to study cases of *Strongyloides* infection in man and autochthonous infections in monkeys, we have found that they also exhibit this same individual relationship.

It is possible that at the time of the migration of preadolescent and adolescent female *Strongyloides* from the lungs to the intestinal tract some of the worms pass down the bowel and out in the feces before they have matured or have had an opportunity to enter the intestinal wall, just as the majority of the parasitic males do. After maturity and penetration of the intestinal mucosa the possibility of such a miscarriage is more remote. As a matter of fact, in many thousands of fecal films of dogs infected with *Strongyloides* which we have examined daily

(5 Gm. samples of feces), during the prepatent, productive and post-productive periods, with a single exception (dog 60), we have never found the slightest evidence of female worms being discharged in the stools. If such a spontaneous discharge of living females from the intestinal mucosa did occur in any appreciable numbers, it might be expected that worms evacuated from higher levels would have an opportunity to reenter the mucosa at lower levels, so that, in the older infections, the larger numbers of worms would be found in successively more distal regions of the intestinal wall. An examination of our data fails to furnish supporting evidence for this assumption. In moderate infections (from 100 to 250 female worms), irrespective of the duration of the infection (dog 62, fourteen days; dog 64, forty-two days; dog 58, eighty-nine days; dog 50, one hundred and twenty days, and dog 48, one hundred and forty days), there was in each case a fairly equal distribution through the duodenum and jejunum, with few worms in the wall of the ileum. In heavier infections (from 400 to 850 female worms) there was a greater tendency than in light infections for a larger number of worms to accumulate in the wall of the posterior jejunum and ileum, although this was likewise not correlated with the duration of the infection (dog 63, forty-one days; dog 60, forty-one days; dog 59, forty-nine days; dog 61, eighty-five days, and dog 41, five months). There were, however, no indications of an actual loss of females in the duodenum being compensated by involvement of the lower levels of the bowel. Thus the reduction in total egg production in any particular infection cannot reasonably be assigned to the evacuation of females from the mucosa. On the contrary, our evidence points to a retention of females within the wall of the dog's intestine for at least several months after the production of eggs has been reduced to zero. A loss of fecundity rather than a loss of female worms, therefore, appears to be the determining factor. What, then, brings parturition to an end?

In our examination of "old" parasitic *Strongyloides* females which are near the end of their productive period we have commonly found 1 or 2 eggs in each horn of the uterus (fig. 2). Such eggs are invariably in the one cell stage and may appear to be well formed, but never proceed with cleavage in utero or after being teased out of the worm. As far as we have been able to test these eggs, they are nonviable. In other females of the same type the few uterine eggs seen are poorly formed, and their contents unorganized. In both of these types there is little, if any, evidence in the ovaries of egg-forming material. In "older" (i. e. postproductive) females, eggs of any description have ceased to be formed; the ovaries are atrophied, and the oviducts and uteri have collapsed. The appearance of all these worms in the tissues

suggests contraction, but when they are removed to physiologic solution of sodium chloride at either 22 or 37 C., they readily elongate and become active. Their digestive tracts are still complete and functional. Except for their sexual functions they appear to be normal. Usually these worms, as they "age," become surrounded by an epithelial cellular capsule, which, in turn, may be encased in a layer of polymorphonuclear leukocytes, or less commonly of eosinophils and monocytes. We have never seen a productive female so encapsulated. It appears, therefore, that reduction in egg production and, finally, complete cessation of this activity are probably due to a local reaction in the tissue of the host.

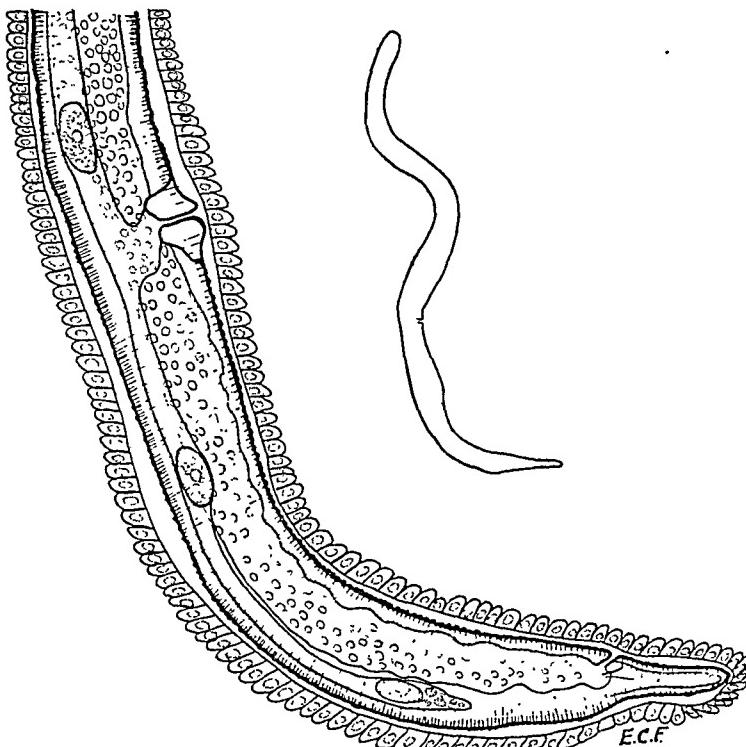


Fig. 2.—Parasitic female *Strongyloides* from the upper duodenum of dog 58 at autopsy on the eighty-ninth day after inoculation. The worm is near the end of the egg-producing period. To the right there is an outline of an entire worm, completely dissected out of the tissues,  $\times 80$ ; to the left, the posterior portion of the worm in its adventitious epithelial capsule is shown. Two apparently fertile eggs may be seen, one in each horn of the uterus. A small amount of egg-forming material is present in the distal end of the posterior ovary. Drawings by means of camera lucida;  $\times 280$ .

This argument is strengthened by additional observations on these "old" females. In some cases the encapsulated worms take on a transparent glossy appearance which makes it difficult to recognize them in films of freshly scraped tissue. Such worms appear to be slowly dying and, on dissection from their adventitious capsules, move sluggishly.

They disintegrate readily under light pressure. Again, worms have been seen from time to time in which the capsule has been partially broken down, with the result that phagocytic cells in the tissue of the host have attacked and partially destroyed one end of the parasite, while the rest of the worm remained alive (fig. 3). These observations suggest the eventual destruction of the complete worm *in situ*. While this type of local reaction of the tissue as a protective mechanism for the host is not unique, it is far less rapid in strongyloidosis than in trichinosis or cysticercosis (*cellulosae*). This milder, slower reaction is probably associated with the lower toxicity of the female *Strongyloides*.

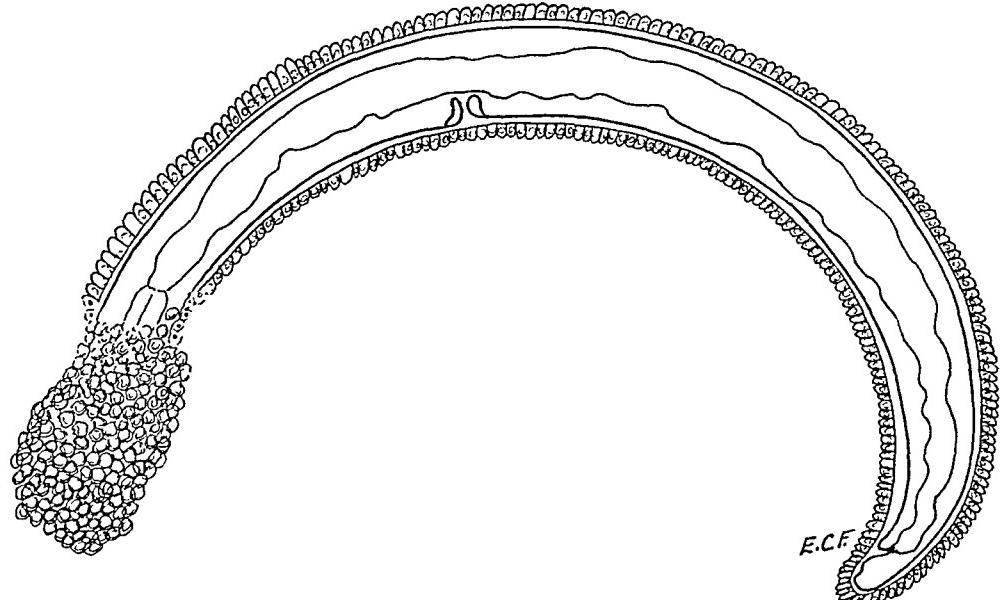


Fig. 3.—Postproductive female *Strongyloides*, within epithelial capsule. All the sexual organs except the vulvar sphincter have degenerated. A group of macrophages has broken down the capsule at the anterior end of the worm and phagocytosed almost the entire esophageal third of the worm. This is the only worm recovered from dog 8 (not in this series), six and a half months after inoculation. The last larvae in the feces were seen three months previously. Drawing by means of camera lucida;  $\times 200$ .

The presence of postproductive parasitic females in the intestinal wall of the host materially complicates the question of specific diagnosis and the correct analysis of the clinical picture. Within the past five years one of us (E. C. F.) has seen twenty-five human cases which did not show a *Strongyloides* infection in examination of three fecal films but in which a single rhabditiform larva was recovered after complete examination of 5 gm. of feces which had been carefully concentrated. In three additional cases clinical and hematologic studies suggested strongyloidosis, although larvae were never recovered in stool concen-

brates. If our canine studies may be used as a criterion, we may expect that either or both of these human types may harbor from several dozen to several hundred postproductive parasitic females, the toxins from which are gradually being absorbed into the system, producing the vague toxic symptoms so characteristic of infection with *Strongyloides*—frequently a leukopenia and a mild eosinophilia, with or without erythropenia. We are convinced that in many cases in which no organisms have been found a careful prolonged search of at least one 5 Gm. specimen of freshly passed feces will result in the discovery of a few larvae of *Strongyloides*. It is equally important to remember that the number of larvae found by fecal examination may not be a criterion of the number of female worms embedded in the intestinal mucosa.

If internal infection occurs in the course of a few months in experimental canine strongyloidosis of human origin, increasing the number of female worms present in the intestinal wall and producing a group of young fertile females, it is altogether likely that over a period of years the same mechanism is at work in man, who is the most suitable host for the human strains of the organism. Among persons of middle age with a history of strongyloidosis of from fifteen to twenty years' standing, there are some who have apparently not been reinfected from the soil since childhood or adolescence. Some of these persons are seriously ill, as indicated by their neurotic behavior and profound malaise. Their blood may or may not show an eosinophilia, but there is usually a leukopenia. They have the signs of a slowly operating toxic process. They are usually constipated but have digestive upsets on the slightest dietary indiscretions, following which there is marked diarrhea, with the passing of mucus in the unformed feces. Examination of the stool may reveal a few rhabditiform larvae of *Strongyloides*; rarely will any considerable number be found. In view of our observations on canine strongyloidosis we are inclined to doubt that in man the female parasite can remain fertile for as long as from fifteen to twenty years. We, therefore, suggest that in such cases the parasites may have been maintained by internal reinfection, even though at the time of examination there is no evidence of dwarf filariform larvae in the feces. Certainly such an explanation has much in its favor and clarifies many of the difficulties which have surrounded these difficult cases of long continued human strongyloidosis. However, one must not overlook the pulmonary focus of adult parasitic female *Strongyloides* as a secondary location from which reinfection may be initiated.

## CONCLUSIONS

Series of controlled experiments on dogs and on a rhesus monkey, undertaken to study the differences in fecundity of parasitic female *Strongyloides*, and, if possible, to determine the cause of these differences, have indicated that the experimental host may be inoculated with equal success when the filariform larvae in the infective stage are introduced (1) cutaneously on the abdominal wall, (2) orally or (3) intracecally.

After the prepatent period the number of eggs produced by the female worms rises rapidly to a peak, declines to a lower level which is maintained for a period of time and then descends lower and lower, finally reaching a base level of zero.

At the beginning of the egg-laying period, or shortly thereafter, the parasitic females invade the intestinal mucosa, usually in the region of the duodenum or jejunum, where they deposit their eggs, ordinarily quite well developed, into the tissues. The eggs usually hatch, and the rhabditiform larvae gradually work their way out into the lumen of the bowel, pass down and are evacuated in the stools. (In simian infections eggs are more commonly passed in the feces and hatch later.) In canine infections some larvae become encapsulated in the mucosa.

Random fecal samplings and culture methods are unsatisfactory indexes of the number of eggs or larvae expelled in the feces. Daily counts of 5 Gm. samples of feces, from which total daily discharges of larvae may be calculated, have been found to be a more reliable measure of such discharges. Great care must be exercised in identifying the larvae, since as many as 90 per cent may be dead or disintegrating at the time of evacuation. Even with these precautions there is a marked daily fluctuation in the counts; this does not appear to be correlated with the actual egg production of the female worms. The average of counts over a period of a week or more, therefore, provides an approximate estimate of the level of production rather than an accurate gage of the productivity of the females.

Daily counts and averages of weekly counts of larvae (or eggs) in the feces are usually far in excess of the number of uterine eggs in the parasitic females, indicating that, on the average, the worms produce from two to four times as many eggs as can be accommodated in their uteri at any one time.

The yield of parasitic females at autopsy is frequently in excess of expectations based on the number of filariform larvae in the inocula. In two experimental animals these mother worms actually exceeded the number of larvae in the infective mediums to which these animals were exposed. This excess can be explained only by the mechanism of internal infection (hyperinfection), by which daughter rhabditiform

larvae transform into dwarf filariform larvae as they pass down the bowel, enter the blood stream through the mucosa of the large bowel, and effect a migration through the lungs and thence to the upper intestinal tract via the respiratory passages, glottis, esophagus and stomach.

Many of the female worms recovered at autopsy were at or near the end of their period of fecundity. In several of these infections larvae had not been recovered from the feces for several weeks or months preceding the death of the host. Such worms were still alive and became active on removal to physiologic solution of sodium chloride, but showed concrete evidence of the termination of the egg-laying period. Many of these worms in the intestinal wall were encapsulated and some were surrounded by layers of white cells, primarily polymorphonuclear leukocytes. In a few instances the capsules had been broken, and the worms were in the process of being phagocytosed.

The reduction in egg production and the eventual loss of adult female worms by the host is not due to the migration of the organisms out of the intestinal mucosa and to their being discharged in the feces, but to encapsulation and phagocytosis by the cells in the tissue of the host, a process comparable in type but not in rapidity to the reaction of the cells to *Trichinella* larvae or to *Cysticercus cellulosae*. It is suggested that this difference is due to the lower toxicity of *Strongyloides*.

Infection of dogs with human strains of *Strongyloides* is maintained for only a period of weeks or months, as contrasted with a period of years in man, the natural host. The period of infection in the monkey lies between these two extremes. Nevertheless, unless internal infection is predicated, it is inconceivable that man remains infected for a period of from fifteen to twenty years without outside exposure or hyperinfection.

In considering the applications of this study to human strongyloidosis, it may be concluded that: (1) there is a need for prolonged intensive fecal examination in suspected cases before the patient is pronounced free from organisms; (2) even when no evidence of organisms is found, such patients may harbor in the intestinal wall several tens or hundreds of postproductive females which may be responsible for characteristic toxic symptoms of chronic strongyloidosis; (3) the number of larvae present in the stools of patients with strongyloidosis is not necessarily an index of the severity of their infection, and (4) internal infection (hyperinfection) is the only satisfactory explanation for prolonged chronic strongyloidosis.

#### SUMMARY

On the basis of an intensive experimental study of human *Strongyloides* in young dogs and of a chimpanzee strain of the organism in a rhesus monkey, concrete evidence has been obtained, indicating that following the period of incubation the parasitic female worms produce

eggs, the number of which rapidly increases and then gradually decreases to zero. This phenomenon is due not to the escape of the worms from the mucosa of the upper levels of the small bowel, but to reactions in the tissues of the host, including first encapsulation of the egg-laying females and later cellular infiltration around, and phagocytosis of, the worms. Ordinary fecal examination for larvae of *Strongyloides* has been found a very unsatisfactory criterion of the presence or numbers of parasitic females, in view of the frequent disintegration of larvae en transit down the bowel and because of the gradual reduction in the egg production of the mother worms. Although fecal examinations may consistently fail to disclose the organism for a period of weeks or months, a considerable number of female worms may still be present in the duodenal and jejunal mucosa and be responsible for chronic toxic manifestations. Internal infection (hyperinfection) is offered as an explanation for prolonged human strongyloidosis.

# NECROSIS OF THE MYOCARDIUM INDUCED BY THE ORTHOPHOSPHATES

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In an earlier publication<sup>1</sup> it was shown that destructive lesions produced by injections of parathyroid hormone are toxic and not secondary to circulatory disturbances. The ultimate mechanism of the toxic action of the hormone was not explained, but because of the characteristic effect of the hormone on the calcium and phosphorus metabolism it was thought that a serious disturbance in one or the other might be the indirect, if not the direct, cause of the necrosis observed. The purpose of the experiments reported here was to study the effects of injected orthophosphates under conditions that prevented their rapid elimination and hence favored their accumulation in the blood.

Whole human blood was found by Kay and Byrom<sup>2</sup> to contain an average of 38.4 mg. of phosphorus per hundred cubic centimeters. Of this amount, 2.9 mg. was in the form of inorganic phosphate. In health the fluctuations from these values were not large. Addis, Meyers and Bayer<sup>3</sup> reported that the concentration of inorganic phosphate in the blood plasma was about proportional to the rate of urinary excretion, and that the kidneys quickly responded to changes in the ingestion of phosphorus phosphate. These authors caused both the concentration of the plasma and the urinary output to rise by injecting a neutral solution of sodium phosphate (75 mg. of phosphorus per kilogram of body weight). Within from two to three hours after the injection the concentration of the plasma had fallen to near normal, as the phosphorus was excreted by the kidneys. Binger<sup>4</sup> injected into dogs amounts of orthophosphates equivalent to 150 mg. per kilogram of body weight. The serum calcium dropped from its normal level of 10 mg. per hundred cubic centimeters to approximately 6 mg. Binger stated that at that

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1. McJunkin, F. A.; Tweedy, W. R., and Brenhaus, C.: Arch. Path. **14**: 649, 1932.
2. Kay, H. D., and Byrom, F. B.: Brit. J. Exper. Path. **8**:240, 1927.
3. Addis, T.; Meyers, B. A., and Bayer, L.: Am. J. Physiol. **72**:125, 1925.
4. Binger, C. A. L.: J. Pharmacol. & Exper. Therap. **10**:105, 1917.

level a condition of tetany supervened, provided that the neutral or alkaline salts had been injected. With acid phosphate solutions the drop in calcium occurred unaccompanied by tetany.

In our experiments we stopped the rapid outflow of injected phosphates by performing bilateral nephrectomy. Special attention was given the cardiac muscle to determine the histologic effects of the injections of orthophosphates, and chemical estimations of the phosphate phosphorus in the blood were made in order to determine whether the incidence of necrosis could be attributed to increased amounts of phosphates in the blood.

#### METHODS

*Histologic Methods.*—Kidneys of young albino rats were removed after ligation of the ureters and vessels at the hilus. The abdominal wounds were closely sutured and sealed with collodion and gauze. All the animals were killed when the experiments were terminated, and the tissues were immediately placed in formaldehyde for fixation. Our procedure was to examine the tissues for necrosis and not for lesser degenerative changes. For this purpose paraffin sections were made and stained with hematoxylin and eosin.

*Chemical Methods.*—The inorganic and total phosphorus of the whole blood were determined by the method of Fiske and Subbarow<sup>5</sup> as modified by Hauch and Koch<sup>6</sup> for small quantities of blood. Blood serum calcium was determined by the Kramer and Tisdall method<sup>7</sup> as modified by Tweedy and Koch.<sup>8</sup> Dried heart muscle was wet ashed with sulphuric acid and hydrogen dioxide and analyzed for total phosphorus by the methods already described.

#### EFFECTS OF BILATERAL NEPHRECTOMY ON INORGANIC PHOSPHORUS OF THE BLOOD AND MUSCLE OF THE HEART

During the first hour following nephrectomy the inorganic phosphorus of the whole blood had risen slightly to 6.5 mg. per hundred cubic centimeters, as compared with an average value of 5.1 mg. in seven normal rats (footnote, table 2). After six hours a definite increase in inorganic phosphorus had appeared (rat 2, table 1). Within eight hours it had increased further to 13.1 mg. and at the end of twelve hours to 13.9 mg. At the end of twenty-four hours there was an average value of 17.7 mg., and after forty-eight the average value was 21.7 mg.

The hearts of the animals were examined histologically for necrosis, but none was observed, except in rats killed forty-eight hours after nephrectomy. In one of these (rat 12) necrosis was distinct. Less severe degenerative changes were not determined by the technic used.

5. Fiske, C. H., and Subbarow, Y.: *J. Biol. Chem.* **66**:375, 1925.

6. Hauch, J., and Koch, F. C.: Unpublished data, personal communication to the authors.

7. Kramer, B., and Tisdall, F. F.: *J. Biol. Chem.* **47**:475, 1921.

8. Tweedy, W. R., and Koch, F. C.: *J. Lab. & Clin. Med.* **14**:747, 1929.

EFFECTS OF BILATERAL NEPHRECTOMY AND INJECTIONS OF DISODIUM HYDROGEN PHOSPHATE

It at once became clear that the reaction of the nephrectomized rat to the injection of phosphate was unlike that of the normal animal. Small doses of phosphate, which were nontoxic for normal rats, pro-

TABLE 1.—*Effects of Bilateral Nephrectomy\**

Rat	Weight, Gm., Individual or Average	Hours After Nephrectomy	Blood Inorganic Phosphorus, Mg. per 100 Cc., Individual or Average	Necrosis of Myocardium
1.....	124	1	6.5	0
2.....	106	6	7.8	0
3.....	120	8	13.1	0
4-5.....	90	12	13.9	0
6-9.....	186	24	17.7	0
10-12.....	133	48	21.7	+ or doubtful

\* In all the tables + indicates single necrotic fibers, ++ intermediate degrees of necrosis and +++ necrotic areas.

TABLE 2.—*Effects of Bilateral Nephrectomy and Intraperitoneal Injections of Solution of Disodium Hydrogen Phosphate*

Rat	Weight, Gm., Individual or Average	Hours After Nephrectomy	Phosphorus Injected,* Individual or Average	Blood Inorganic Phosphorus, Mg. per 100 Cc., Individual or Average	Necrosis of Myocardium
1-3	117	48	2.6	19.8	++
4-6	248	48	4.4	20.6	Doubtful
7-10	178	24	3.4	21.4	Doubtful
11-12	119	7	2.7	11.0	0
13-14	130	3	2.9	6.7	0
15	120	1½	2.8	8.3	
16	133	¾	3.6	5.9	
17-19	144	24	6.8	...	++
22-22	112	24	12.8	30.9	+++
23-25	116	24	3.2	12.8†	
26-31	114	24	3.2	14.2†	
32-37	121	4¼	32.3	53.3	

\* Injections were made intraperitoneally immediately following closure of the abdominal incision. A solution of disodium phosphate, which contained 0.75 Gm. of the salt per hundred cubic centimeters, was diluted appropriately before injection.

† The average total blood phosphorus for rats 23 to 25 was 85.2 mg. A total blood phosphorus of 51.3 mg. was the average in seven normal rats of an average weight of 132 Gm., and in the same normal animals the inorganic phosphorus had an average of 5.1 mg. In six additional nephrectomized rats which received injections (rats 26 to 31) the average total blood phosphorus was 88.4 mg.

duced destructive lesions in the nephrectomized animals, and larger doses, while nontoxic for normal rats, were lethal for the nephrectomized rat. Rats 1 to 3, although given injections of small doses of the basic disodium hydrogen phosphate, showed much myocardial necrosis. In the larger animals the amount of necrosis was small or absent, and later in the work it was found that rats which weighed about 100 Gm. were most susceptible to the cardiac injury. Rats which weighed more than 100 Gm. were resistant and showed the necrosis irregularly. In

severe lesions, such as that seen in rat 1 (table 2), the necrotic areas were of sufficient size to be seen on gross examination as opaque foci in the ventricular walls.

The lesions were in proximity to the coronary vessels. Necrosis also appeared in the media of the first part of the aorta and was occasionally seen in the walls of the larger coronary arteries. The earlier of the lesions appeared microscopically as myomalacia, while at the end of forty-eight hours many leukocytes had accumulated about the necrosis, which made the focus much more distinct.

Deposition of calcium in both the cardiac and the aortic lesions was observed. In the striated musculature of the abdominal wall necrosis was seen, but the lesions of the myocardium were the most severe. Repeated examination of the liver, lungs and suprarenal glands failed to show destructive lesions. In the wall of the stomach beneath the muscularis mucosae there was often much edema.

It is clearly seen in table 2 that the necrosis resulted from the combined effect of the nephrectomy and the injected phosphate and that doses of from 6.8 to 12.8 mg. (rats 17 to 22) regularly produced the myocardial injury.

The many determinations of blood inorganic phosphorus and total phosphorus which were made from twelve to twenty-four hours after nephrectomy and the injection of small doses of phosphate showed only a doubtful relationship of either of these values to the incidence of myocardial necrosis. On the other hand, nephrectomized rats which received injections of doses sufficiently large to produce regularly the myocardial lesions showed an average inorganic phosphorus of 30.6 mg. at the end of twenty-four hours (rats 20 to 22, table 2), as compared with a value of 17.7 mg. in nephrectomized rats not given injections (rats 6 to 9, table 1). The movement of larger doses of phosphates injected intraperitoneally into the blood stream was shown in a striking fashion, since by this experiment the blood inorganic phosphorus was increased in six rats (32 to 37, table 2) to an average of 53.3 mg. within an average time of four and a quarter hours. Normal rats given injections of comparable doses showed no ill effects, and the blood inorganic phosphorus in three normal rats was elevated to an average of only 9.4 mg. After large injections into normal rats no lesions were seen in the heart on histologic examination.

#### EFFECTS OF BILATERAL NEPHRECTOMY AND INJECTIONS OF OTHER ORTHOPHOSPHATES

It was thought that the moderate tendency of the basic disodium hydrogen phosphate to produce alkalinization might influence the reactions resulting from the administration of the salt. In rats 1 to 3

(table 3), weighing from 176 to 200 Gm., which received small doses of a solution of disodium and monopotassium phosphates buffered to  $p_H$  7.1, the inorganic phosphorus of the blood was approximately the same as in rats receiving the basic disodium hydrogen phosphate alone, and there was no necrosis of the myocardium. When the buffered solution was injected into smaller rats (4 to 7) a slight amount of necrosis was seen.

Since it has been determined that tetany in the dog is not produced by injections of the monosodium phosphate, the effect of this salt on the cardiac muscle was investigated. In rats 8 to 14 (table 3) it appears that this acid phosphate did not affect the heart so much as the disodium salt, and large doses (rats 15 to 17) caused less severe lesions than comparable doses of disodium hydrogen phosphate. Rats given injec-

TABLE 3.—*Effects of Bilateral Nephrectomy and Injections of Monosodium Phosphate, Disodium Phosphate, Trisodium Phosphate and Di-Ammonium Phosphate*

Rat	Weight, Gm.	Hours After Nephrec- tomy	Phos- phorus Injected, Mg.	Type of Solution Injected	Blood Inorganic Phosphorus, Mg. per 100 Cc.	Necrosis of Myocar- dium
1-3	188	24	6.7	Buffer 1*	21.6	0
4-7	131	24	5.5	Buffer 2*	15.3	+
8-10	159	24	4.3	Monosodium phosphate	15.2	0
11-14	123	48	4.8	Monosodium phosphate	....	0
15-17	125	24	11.3	Monosodium phosphate	25.5	++
18-19	129	24	20.0	Sodium carbonate	....	0
20-21	99	24	50.0	Sodium chloride	....	0
22-26	88	27	9.6	Di-ammonium phosphate	38.1	0

\* Buffers 1 and 2 were mixtures of disodium hydrogen phosphate and potassium dihydrogen phosphate buffered to  $p_H$  7.1 and 7.2, respectively. Rats 5 and 6 also received a subcutaneous injection of 2 cc. of a 0.5 per cent solution of sodium carbonate. Rats 18 and 19 received sodium carbonate alone. Rats 20 and 21 were given injections of sodium chloride alone.

tions of varying doses of di-ammonium phosphate (rats 22 to 26) showed no destructive myocardial lesion. The small doses of trisodium phosphate which were injected produced no change in the heart. A buffered solution of phosphate when combined with sodium carbonate injected subcutaneously (rats 5 and 6, table 3) revealed no increase of myocardial damage. Indications that the sodium ion was not responsible for the myocardial necrosis were obtained by injections of sodium chloride (rats 20 and 21). Sodium carbonate was also without effect (rats 18 and 19).

#### RELATIONSHIP OF MYOCARDIAL NECROSIS TO SERUM CALCIUM AND INJECTIONS OF CALCIUM GLUCONATE

Up to this point it appeared that the lesion in the heart was caused directly or indirectly by the phosphate ion of the injected phosphate and that its production was favored by the use of the basic salt. We

were impressed by the tendency of the lesions to calcify and were led to investigate the calcium content of the serum and the effect of injections of calcium gluconate.

In view of the well known evidence indicating the mutual control of the concentration of calcium and phosphate ions in the blood plasma, it was thought that a rapid rise in the blood inorganic phosphate induced by nephrectomy or nephrectomy plus injected phosphates might be reflected in a value for serum calcium well below normal. The serum calcium was determined in two nephrectomized rats (8 and 9, table 4) which had been given injections of about 5 mg. of phosphate phosphorus. About twenty-four hours later, the serum calcium was 10.8 and 11.4 mg. per hundred cubic centimeters, respectively, while the phosphorus had risen to 17.8 and 16.0 mg., respectively. Then

TABLE 4.—*Serum Calcium After Bilateral Nephrectomy and Injections of Disodium Phosphate*

Rat	Average Weight, Gm.	Hours After Nephrectomy	Phosphorus Injected, Mg. per 100 Cc., Individual or Average	Serum Calcium, Mg. per 100 Cc., Individual or Average	Blood Inorganic Phosphorus, Mg. per 100 Cc., Individual or Average
1-3	162	24	0	8.95	
4-5	213	3½	58.2	8.08	
6-7	147	1¼	40.1	8.22	
8-9	208	21	4.7	11.10	16.9
10	220	72	2.8	11.80	20.3
11-12	178	24	19.2	6.73	
13-14	199	4	21.1	9.24	
15-16	158	2	16.6	8.63	
17-18	202	16	21.0	8.53	
19-20	190	6	20.0	9.28	
21-22	204	12	22.3	6.73	
23-25	214	9	22.8	7.85	

rats 11 to 25 were given injections of an amount of phosphorus that produced regularly myocardial necrosis. At the end of from eight to twelve hours a slightly lowered calcium level was observed, and after twenty-four hours there was pronounced hypocalcemia. Large doses of phosphate (rats 4 to 7, table 4) lowered the calcium level only moderately within from one and a quarter to three and a half hours. At the end of twelve hours, however, a dose of 22.3 mg. caused marked hypocalcemia (rats 21 and 22).

These results show that even in nephrectomized rats it is necessary to use an amount of orthophosphate greatly in excess of the toxic dose in order to demonstrate a sudden lowering of the serum calcium. Several experiments made in connection with another investigation demonstrated that the serum inorganic phosphorus may reach a value of three or four times its normal concentration as a result of nephrectomy, while the serum total calcium may either remain within its normal range or fall to not more than 10 per cent below the average total

serum calcium of normal rats. As the chemical methods used in this investigation measured total amounts of serum inorganic phosphorus and calcium and since no reliable methods for the measurement of the concentration of the calcium and phosphate ions were applicable, it is by no means certain that the relationship of these ions was undisturbed.

Collip<sup>9</sup> stated that lesions could not be produced by injections of calcium salts alone. However, by the injection of huge amounts (400 mg.) of calcium gluconate into normal small rats weighing less than 150 Gm. we<sup>1</sup> were able to produce small foci of necrosis in the kidneys. In the nephrectomized rat an intraperitoneal dose of about 75 mg. of the gluconate was sufficient to cause severe destructive lesions in the myocardium and the aortic media (table 5). In both locations the necrotic areas showed within forty-eight hours the same tendency to calcify observed in the "phosphate" lesions.

TABLE 5.—Effect of Bilateral Nephrectomy and Injections of Calcium Gluconate

Rat	Average Weight, Gm.	Hours After Nephrectomy	Calcium Gluconate Injected, Mg. per 100 Cc.	Serum Calcium, Mg. per 100 Cc.	Blood Inorganic Phosphorus, Mg. per 100 Cc.	Myocardial Necrosis
1	284	24	75	9.51	...	++
2	276	24	75	10.36	37.5	+
3	288	24	60	9.62	20.8	+
4	282	24	85	19.40	19.0	
5	335	24	95	15.14	12.6	
6	285	24	65	10.00	16.0	+++
7	300	24	80	10.40	21.8	+++
8	310	72	110	.....	....	++*

\* There was also much calcification in the media of the coronary artery, as shown by staining with both hematoxylin and eosin.

In only two instances (rats 4 and 5, table 5) was there pronounced hypercalcemia. On the other hand, the inorganic phosphorus reached an average concentration well above that found in nephrectomized rats which did not receive injections. Inability of the excess calcium to hold back the mounting phosphorus level permitted this condition of both hyperphosphatemia and hypercalcemia to develop in the blood at the end of the twenty-four hour period.

#### COMMENT AND SUMMARY

It is recognized that with the cessation of renal function there ensues a variety of progressive chemical changes in the organism. In discussing the effects of ligation of the ureters in the dog, Atchley and Benedict<sup>10</sup> stated:

Profound changes occur in the distribution of the electrolytes of the blood and tissues. The net effect on the blood serum is, in résumé, a retention of

9. Collip, J. B.: Am. J. Physiol. **76**:742, 1926.

10. Atchley, D. W., and Benedict, E. M.: J. Biol. Chem. **73**:1, 1927.

phosphate and sulphate which takes base from carbonate and chloride. This equimolecular interchange occurs regardless of the movement of water to or from the tissues, and seems to have no effect on the total base content.

It is evident that as a result of these changes an ultimate condition of acidosis supervenes. The data presented here show that it is approximately forty-eight hours after nephrectomy, when the aforementioned changes are probably well advanced, that histologic evidence of beginning necrosis may appear in some animals. However, injection of orthophosphates into the rat immediately after nephrectomy in quantities that do not kill or produce histologic lesions in the normal rat injured the cardiac muscle to such an extent that necrosis appeared regularly at the end of twenty-four hours in the animals studied. In connection with our experimental data, it has been pointed out that the lesions are produced in the nephrectomized animal with either alkalosis or some degree of acidosis such as may be produced by the dihydrogen phosphate. That alkalosis rather than acidosis favors the production of the lesion is probably indicated by the failure of di-ammonium phosphate to produce myocardial necrosis. In the varied chemical analyses made we obtained no direct evidence that the injected orthophosphates actually entered the cells of the myocardium and initiated the changes responsible for the necrosis observed. Determinations made on the desiccated hearts of nephrectomized rats which were given injections showed no greater total phosphorus than in the cardiac muscle of normal rats. Application of the Fiske-Subbarow-Mallory method<sup>11</sup> to pyroxylin (celloidin) sections prepared from the hearts with the "phosphate" lesions showed no microscopic evidence of excess phosphorus.

Early calcification of the "phosphate" lesions was frequently seen. Our experimental data reveal hyperphosphatemia at the time that the myocardial necrosis was developing. With an excess of phosphate ions in the part, there would be a tendency toward precipitation of calcium entering the area or already in it. The similarity of the "phosphate" lesions and lesions caused by injections of calcium gluconate leads one to suspect a similarity in their pathogenesis. Ham and Lewis,<sup>12</sup> in a study of lesions caused by viosterol, regarded the deposition of calcium as the primary causative factor in their production. In our investigation<sup>1</sup> of lesions caused by parathyroid hormone it was made clear that in no instance was calcification observed, except in association with destructive tissue changes. Since calcification in the microscopic sense represents a great absolute increase of calcium in the tissues affected, it may well be that excess calcium was thrown out of solution for a considerable time before its histologic appearance. Other types of injury

11. Mallory, F. B.: Am. J. Path. 9:557, 1933.

12. Ham, A. W., and Lewis, M. D.: Arch. Path. 17:356, 1934.

to the myocardium of the rat apparently manifest little affinity for calcium salts. The heart was "snagged" by means of syringe needles and examined at twenty-four, forty-eight and seventy-two hour intervals. Although conspicuous necrotic foci were present along the needle tract, no trace of calcification was present.

#### CONCLUSIONS

Myocardial necrosis may be produced by the injection of disodium phosphate into small nephrectomized rats. Lesions closely resembling the "phosphate" necrosis may be produced by the injection of calcium gluconate into nephrectomized rats. Both kinds of lesions show a tendency to early calcification. The chemical and histologic evidence indicates a close relationship between the necrosis and the deposition of calcium. In view of the tendency of the myocardial necrosis to become calcified it may be assumed that the precipitation of calcium and phosphate ions represents a local condition of tissue in which the highly organized parenchymal cells of the myocardium cannot survive.

# CONCURRENT TUMORS OF THE LEFT CAROTID BODY AND BOTH ZUCKERKANDL BODIES

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ROCHESTER, MINN.

The carotid body was first described by von Haller,<sup>1</sup> in 1743, and again by Neuber,<sup>2</sup> in 1783. However, little attention was paid to their work, and it remained for the detailed description of Luschka,<sup>1</sup> in 1862, to stimulate genuine interest. Marchand,<sup>3</sup> in 1891, was the first to describe a tumor of the carotid body, and Paltauf,<sup>3</sup> in 1892, was the second. Since that time a great number of reports of tumors of these bodies have appeared in the literature. The reports have been extensively reviewed in the past six years by Talman,<sup>1</sup> Bevan and McCarthy<sup>4</sup> and Rankin and Wellbrock.<sup>5</sup> The investigators last mentioned recorded one hundred and ninety-six cases. Thus, it is apparent that tumors of these glands are not rare, even though they may have been reported infrequently.

In 1901, Zuckerkandl<sup>6</sup> discovered and described in detail, two small bodies which lie on the anterior surface of the abdominal aorta, one on each side of the point of origin of the inferior mesenteric artery. He found that these small bodies were consistently present and easily visible in all human fetuses from 5 months of age to full term, but that after this period they atrophied rapidly, so that at 2 years of age they were difficult to find and by the tenth year they could be found only by serial sectioning of the tissue in the regions in which they were known to occur in early life. He further noted that the bodies possessed the same histologic appearance as that of the carotid body and suprarenal medulla. The minute periaortic masses have since been known as Zuckerkandl bodies or glands.

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This work was done under the direction of Dr. John deJ. Pemberton, Division of Surgery, the Mayo Clinic.

1. Quoted by Rankin and Wellbrock.<sup>5</sup>
2. Neuber, quoted by Bevan and McCarthy.<sup>4</sup>
3. Quoted by Rabin.<sup>22</sup>
4. Bevan, A. D., and McCarthy, E. R.: Surg., Gynec. & Obst. **49**:764, 1929.
5. Rankin, F. W., and Wellbrock, W. L. A.: Ann. Surg. **93**:801, 1931.
6. Zuckerkandl, E.: (a) Verhandl. d. deutsch. anat. Gesellsch. **15**:95, 1901. (b) The Development of the Chromaffin Organs and of the Suprarenal Glands, in Kiebel, Franz, and Mall, F. P.: Manual of Human Embryology, Philadelphia, J. B. Lippincott Company, 1912, vol. 2, p. 157.

In 1865, Henle discovered that in the suprarenal gland, fixed in a solution containing a chromium salt or acid, a yellowish-brown pigmentation, which he described as granular, developed frequently in the medullary cells. This fact was confirmed by Soulié,<sup>7</sup> Kohn,<sup>8</sup> Wiesel<sup>7</sup> and many others, and was called the chromaffin reaction, because of the affinity of the medullary tissue for the chromium salts. Stilling,<sup>9</sup> in 1892, was the first to demonstrate the presence of this reaction in cells of the carotid body; this was confirmed by Kohn and Zuckerkandl and many of the men who later described tumors of these bodies. Zuckerkandl demonstrated the same reaction in the periaortic bodies which bear his name. Thus he, Kohn and others believed that these tissues were of common origin and together composed the chromaffin system.

Since Stangl<sup>10</sup> first described a tumor of a Zuckerkandl body, in 1902, I have been able to find reports of only three others in the literature; no other case of concurrent tumors of a carotid body and both Zuckerkandl glands has been reported. For this reason the present case should be of particular interest.

#### REPORT OF A CASE

*History.*—A housewife, aged 39, was admitted to the Mayo Clinic on Sept. 13, 1933, complaining of a lump in the left side of her neck. She first noted a small nodule in this region five years previously. For a short time it increased in size, but growth soon became more or less stationary, with only slight fluctuations. There had been no local pain, but since the onset the patient had been annoyed by an almost constant, dull aching pain in the left suboccipital region and in the left side of her neck. Frequent attacks of severe, shooting pain occurred in this region of the neck, extending to the region of the left ear. These attacks lasted only a few seconds, but were often accompanied by a feeling of numbness and dizziness that lasted much longer. The patient's neck was not stiff and she had no difficulty in speaking or swallowing. She had not lost weight. Roentgenologic treatment elsewhere produced no noticeable diminution in the size of the tumor. The only facts of note in her history were that she underwent appendectomy in 1918 and cholecystectomy in 1932.

*Physical Examination.*—A discrete, firm mass, extending forward beneath the sternocleidomastoid muscle, was found on the left side of the neck just below the angle of the jaw. It measured approximately 3.5 cm. in length and 2.5 cm. in width, and was elevated 1.5 cm. above the normal contour of the neck. The skin was freely movable over it, but the mass was bound down to the underlying structures and could be moved only with difficulty. Moderate pressure on the mass elicited slight tenderness. The nose and throat were normal. The general physical condition was good except for a slight degree of hypertension; the blood pressure was 160 mm. of mercury systolic and 86 diastolic. Urinalysis and all

7. Quoted by Zuckerkandl.<sup>6b</sup>

8. Kohn, A., quoted by Eisenberg and Wallerstein,<sup>23</sup> Zuckerkandl,<sup>6</sup> Rabin<sup>22</sup> and Smith.<sup>11</sup>

9. Stilling, quoted by Smith.<sup>11</sup>

10. Stangl, E.: Verhandl. d. deutsch. path. Gesellsch. 5:250, 1902.

examinations of the blood gave normal results. In a roentgenogram of the cervical region, the tumor was seen to lie near the surface of the spinal column and did not appear to be attached to it. The exact nature of the mass was questionable, and three preoperative diagnoses were considered, namely, tumor of the carotid body, neurofibroma and mixed cell tumor.

*Operation and Course.*—A large tumor of the carotid body was found, which measured 5 by 3.5 by 2.5 cm. The common carotid artery and both the internal and the external carotid arteries were surrounded at the point of their bifurcation, so that in removing the mass the common carotid artery had to be ligated and the bifurcation removed. The pathologist reported a malignant tumor of the carotid body with invasion of the capsule.

The immediate postoperative course was satisfactory; no residual signs of injury to the nerves were manifest. However, during the night of the second postoperative day, right hemiplegia developed along with complete aphasia. The following day signs of bronchopneumonia were noted, and death occurred one week after operation.

*Necropsy.*—On examination of the embalmed body shortly after death, the left middle cerebral artery was found to be thrombosed, with infarction of the left parietal lobe of the brain. The operative site was not disturbed. The heart was normal. Both lower lobes of the lungs were covered with large areas of thick, fibrinous exudate and revealed extensive bronchopneumonia. The appendix and gallbladder were absent. The suprarenal glands and all other abdominal viscera appeared normal. On the right anterolateral surface of the abdominal aorta just lateral to the origin of the inferior mesenteric artery, was a firm, nodular, yellowish-white tumor measuring 3 cm. in length, 2 cm. in width and 1.5 cm. in thickness. It was attached to the aorta by thin strands of fibrous tissue, but appeared to be held in position mainly by numerous branches of the sympathetic nerves which passed along the aorta. The nerve strands entered the substance of the tumor and could not be dissected from it. One surface of this body rested against the inferior vena cava, and when the latter was opened two of the nodules of the tumor were seen to be markedly indenting, although not perforating, its wall. On the left anterolateral aspect of the aorta, mostly caudal to the origin of the inferior mesenteric artery, lay another discrete mass similar in shape to the one on the right side. Its surface, however, was smooth, its color yellowish brown and its consistency soft. It would have been considered a large lymph node except for its consistency and the fact that numerous nerve fibers entered its substance, as they entered the tumor on the right. The mass on the left was slightly smaller than the other, measuring 2.5 cm. in length, 1.5 cm. in width and 0.8 cm. in thickness. Lying in the fatty tissue just below the celiac axis were three small, discrete, bright yellow, spherical bodies of fairly firm consistency. They were each 0.4 cm. in diameter. A few fine nerve fibers were seen to extend to them, but these were torn away when the aorta was removed (fig. 1).

The tumor taken from the neck at operation was grayish white and of firm consistency. It had a slightly nodular surface. Its measurements have already been given. Many fragments of nerve strands were hanging from its surface. The common carotid artery entered the lower pole of the tumor and bifurcated in its substance, so that the internal and external carotid arteries emerged from its upper pole. The cut section also presented a firm, yellowish-white surface.

*Histologic Examination.*—Tumor of the Carotid Body: Sections from various parts of the cervical tumor were stained with hematoxylin and eosin. Cellular arrangement and density varied widely. In the majority of sections the cells

were arranged in densely packed masses with no particular distribution. However, in a few areas they were much more loosely joined to form whorls and thick strands; these formations were caused by an interlacing network of fine fibrous septums and capillary blood sinuses. The densely packed cells were oval and polyhedral, and gave the impression of being epithelial. In some of these areas the outlines of cells were indistinct, so that the cells appeared to be fused together into large, multinucleated, syncytial masses. The loosely joined cells were almost entirely stellate, with three or four pseudodendritic processes extending from them in various directions and joining with those of other cells, thus forming a ganglion-

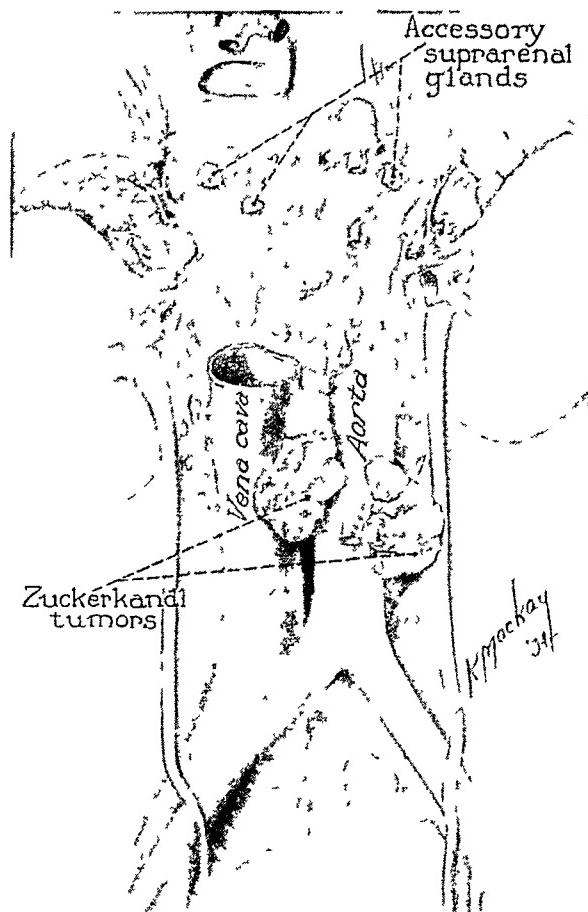


Fig. 1.—Accessory suprarenal glands below the celiac axis and Zuckerlandl tumors on either side of the abdominal aorta.

like network. Between adjoining cell processes in these areas were large, clear spaces, a few of which were lined by endothelium and contained erythrocytes; the remaining clear spaces probably were artefacts due to a shrinkage of cells.

The cytoplasm of the parenchymal cells appeared in two distinct forms: In the larger number it stained homogeneously with eosin, whereas in others it was very pale and contained a deeply staining eosinophilic reticulum. The nuclei varied markedly, in both size and shape; the round and oval types predominated, but there were also a number of large, irregularly shaped nuclei, many of which appeared to be polymorphous. Practically all these nuclei contained hyperchromatic reticulum and not infrequently a small, eccentrically situated nucleolus. Scattered

diffusely and in clumps throughout all sections were large numbers of cells resembling lymphocytes; each consisted of a dark nucleus surrounded by a thin border of cytoplasm. No definite mitotic figures were observed, but the parenchymal cells had invaded the outer capsule in several places (fig. 2A).

Tumors of the Zuckerkandl Bodies: In spite of the fact that the two tumors differed in gross appearance, their microscopic structure was essentially the same. Sections were taken longitudinally through the bodies and also from one of the nodules encroaching on the wall of the inferior vena cava. They were stained with hematoxylin and eosin. A definite fibrous capsule was seen, with small fibrous septums extending from it into the substance of the tumor which divided the tissue into irregular lobules. Large numbers of capillary blood vessels, lined with a single

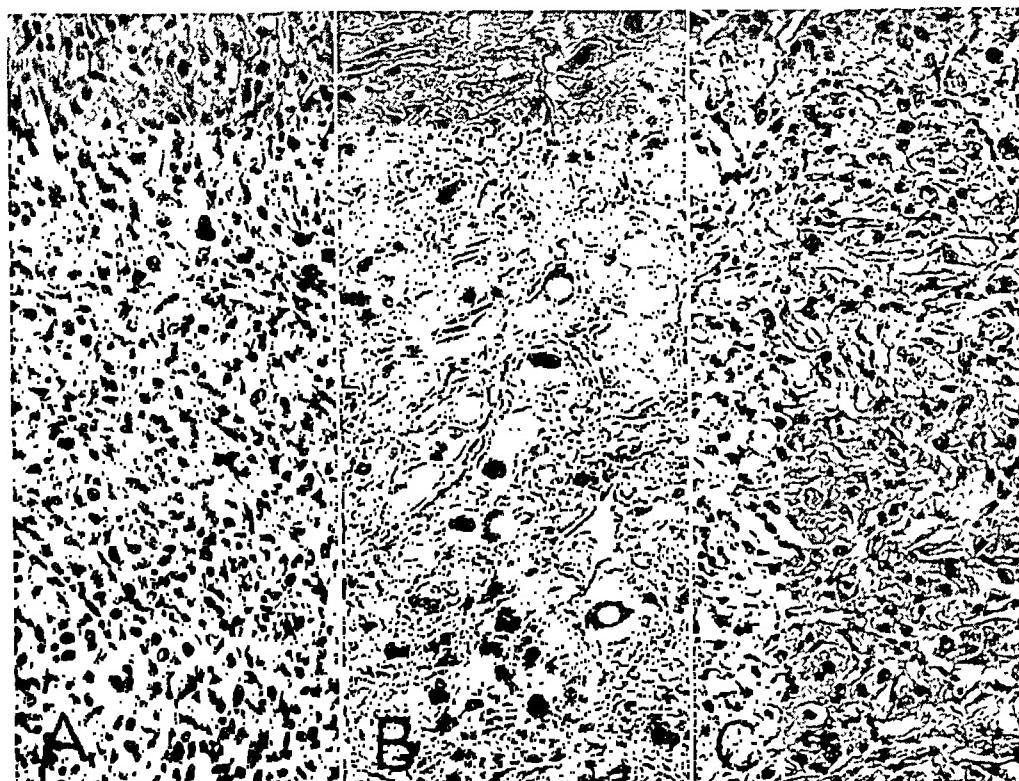


Fig. 2.—*A*, tumor of the left carotid body; *B*, positive chromaffin reaction as demonstrated by the tumor of the right Zuckerkandl body; *C*, tumor of the left Zuckerkandl body.

layer of endothelium and containing erythrocytes, formed an interlacing network. The parenchymal cells exhibited the same features as did those of the cervical tumor, except that the former were densely packed in the peripheral layers and more loosely joined in the central areas. The left periaortic body was less cellular and contained more blood sinuses than did the one on the right, which probably accounted for the fact that it was grossly much softer. The cells had the same polyhedral and stellate shapes, with indefinite outlines and cobweb-like cytoplasm, that were seen in the cervical tumor. However, in those sections fixed in a 5 per cent aqueous solution of potassium dichromate, the cells were clearly outlined and all were oval and polyhedral, the stellate shapes were entirely absent and the cytoplasm stained homogeneously. For these reasons, I believe the stellate

forms to be artefacts produced by the shrinkage of cells. The nuclei of the cells of the Zuckerkandl bodies were round or oval, and corresponded in size to those of the cells in which they were situated. All these nuclei likewise contained hyperchromatic reticulum, and in many there was a small nucleolus. In some, also, was a large, clear vacuole, which gave the nucleus the appearance of a signet-ring. The lymphocyte-like cells found in sections of the cervical tumor were also seen here, although there were comparatively fewer of them; they were distributed separately and in small clumps. An occasional mitotic figure also was observed. The parenchymal cells had invaded the surrounding capsule in several areas and appeared actually to have perforated the wall of the vena cava at one point (fig. 3).



Fig. 3.—Section through the wall of the vena cava, demonstrating the degree of extension of cells of the right Zuckerkandl tumor.

The sections from the three tumors just described were so remarkably similar, histologically, that they could not be distinguished one from the other except by the area of more loosely joined cells in the center of the Zuckerkandl bodies (fig. 2 B and C).

**Accessory Suprarenal Glands:** Sections taken from the center of two of the three small yellow nodules which, as already mentioned, were found attached to the anterior surface of the abdominal aorta above the two Zuckerkandl tumors were composed almost entirely of suprarenal cortex with the typical glomerular layer and the fasciculi converging toward the center. A wide reticular layer also was present, with the usual pigment in many of its cells. Situated eccentrically in one section was a small area of basophilic, stellate cells which resembled normal suprarenal medulla. The nodules were without doubt accessory suprarenal glands; one was complete.

*Tests for Epinephrine.*—The tumor of the carotid body, removed at operation, was preserved for seven days in a solution of formaldehyde, U. S. P. (1:10). From various regions in this mass sections  $\frac{1}{8}$  inch (0.32 cm.) in thickness were taken, and extracts were made by macerating each in 6 cc. of distilled water, precipitating the protein with trichloracetic acid and filtering. Tests for epinephrine were made by adding to half of this filtrate from 2 to 3 minims (123 to 185 mg.) of a 10 per cent solution of potassium ferricyanide and neutralizing with a saturated solution of sodium bicarbonate. Three sections of the tissue tested in this manner gave a negative reaction for epinephrine. Two sections removed from the periaortic tumors were similarly tested and found to contain no epinephrine, even though they had not previously been preserved except for the embalming of the body before necropsy. The other halves of all of the filtrates were tested by the Vulpian reaction, which similarly gave negative results. Unfortunately, the accessory suprarenal bodies were not tested. Sections were taken from the suprarenal glands which contained visibly large amounts of medulla. These sections were analyzed by the same method and proved to give strongly positive reactions for epinephrine. Thus, it is apparent that any appreciable amount of epinephrine in the periaortic tumors at the time of death could not have been entirely destroyed or washed away by embalming.

*Tests for Chromaffinity.*—Sections similar to those used in the previous tests were placed in a 5 per cent aqueous solution of potassium dichromate for three days. They were then mounted in paraffin and stained with hematoxylin. Sections from the tumor of the carotid body were entirely negative, but those from the periaortic bodies exhibited a strongly positive chromaffin reaction. Cells of the suprarenal medulla reacted much more weakly, but were definitely positive.

#### COMMENT

As stated previously, the published reports and reviews on the carotid body and its tumors have covered the field so completely that I wish to mention only the theories of the embryologic origin of the gland in order that there may be a better understanding of the reason for its classification in the chromaffin or paraganglionic system: 1. The theory that the carotid bodies are derived from pharyngeal epithelium has generally been discarded, as the cells of the carotid body have been shown to have no similarity to those derived from the third pharyngeal pouch. Smith<sup>11</sup> demonstrated that the glands were derived from primordial nerve cells in this region and not from epithelium. 2. A few still use the term "perithelioma" for tumors of these organs, on the assumption that the bodies are derived from the perithelium or endothelium of the carotid arteries, but most investigators no longer accept this theory, and Smith's work also disproves it. As early as 1902, both Kohn<sup>12</sup> and Zuckerkandl stated that these bodies "do not arise from thick spots on the arteries." Since perithelial or endothelial cells are probably only flattened fibrocytes, cells of the carotid body could hardly have been derived from them. 3. The third theory is that the bodies are derived

11. Smith, Christianna: Am. J. Anat. 34:87, 1924.

12. Kohn, Alfred: Arch. f. mikr. Anat. 61:81, 1900.

from the "sympathogonia cells," as described by Kohn in 1903. These cells are of ectodermal origin, and are thought to constitute the anlage of both sympathetic and chromaffin systems. Thus, they may develop into mature ganglionic cells of the sympathetic system, or they may differentiate to form the chromaffin cells of carotid bodies, the medulla of suprarenal glands, the Zuckerkandl bodies and other small paraganglions which are said to lie along the course of the abdominal aorta. Most investigators agree that, morphologically, the carotid body and its tumors are characteristic of the paraganglionic system. The positive chromaffin reaction is also typical. However, Vassale,<sup>13</sup> Aszoda and Paunz,<sup>13</sup> DeCastro,<sup>13</sup> Chase<sup>14</sup> and Christie<sup>15</sup> have never been able to demonstrate the presence of epinephrine in these bodies. The histologic appearance of the cervical tumor in my case closely resembled this type of tissue, but, owing to the fact that it was previously preserved in solution of formaldehyde, I do not feel justified in attempting to compare the negative histochemical results with those in other cases.

As was stated in the introduction, Zuckerkandl noted a similarity between the morphologic character and chromaffin reaction of the periaortic body and those of the medulla of the suprarenal glands. He did not refer to the presence of epinephrine, but Handschin<sup>16</sup> stated that Biedl,<sup>17</sup> Wiesel<sup>17</sup> and Danisch<sup>17</sup> found it in extracts of Zuckerkandl bodies removed from fetuses ranging in age from 6 months to full term. Since the time of this work, many investigators have corroborated Zuckerkandl's findings, but the presence of epinephrine is still questionable. The four reports of tumors of this body are interesting, because each tumor presented certain characteristics of the chromaffin system, although none presented all of them.

Stangl<sup>10</sup> reported the case of a man, 32 years of age, who complained of an abdominal mass which he had first noticed three months previously. At operation, a discrete tumor the size of an apple was found resting on the aortic bifurcation. This mass was identified as tumor of a Zuckerkandl body by its histologic appearance and by a strong chromaffin reaction. No tests were made for epinephrine; the patient's blood pressure remained normal, as it had before operation.

Hausmann and Getzowa<sup>18</sup> (1922) on postmortem examination of the body of a man 54 years of age, who had died of pneumonia, found, incidentally, a tumor the size of a hen's egg attached to the aortic bifur-

13. Quoted by Christie.<sup>15</sup>

14. Chase, W. H.: J. Path. & Bact. **36**:1, 1933.

15. Christie, R. V.: Endocrinology **17**:421, (July-Aug.) 1933.

16. Handschin, Erna: Beitr. z. path. Anat. u. z. allg. Path. **79**:728, 1927.

17. Quoted by Handschin.<sup>16</sup>

18. Hausmann, Max, and Getzowa, Sophie: Schweiz. med. Wchnschr. **52**:911, 1922.

cation. Its histologic appearance was characteristic of paraganglionic tissue, and it also exhibited a strong chromaffin reaction. No actual tests were made to determine the presence of epinephrine but, because the patient had been seen over a period of four years and had shown signs of nervousness, tremor, profuse sweating and pulmonary bleeding, and because a low value for blood sugar had been found on last admission in addition to the findings at necropsy of an enlarged heart and enlarged kidneys, Hausmann and Getzowa felt that epinephrine probably had been produced by the tumor during the patient's life.

Handschin (1928) described the case of a man, 45 years of age, who had died of carcinoma of the stomach. At necropsy, a tumor the size of a plum was discovered at the aortic bifurcation. It was histologically similar to the chromaffin tumors previously described, but gave no chromaffin reaction, even though the tissue was placed in fixative as short a time as three hours after death. However, an extract of the tissue dilated the pupil of an isolated frog's eye, and this was considered a positive test of epinephrine.

Nordmann and Lebküchner<sup>19</sup> (1931) reported two cases of paraganglionic tumors; only one of these, however, can be classed definitely in this series. A man, 53 years of age, had been killed in an accident. At necropsy, a tumor the size of a kidney was found at the aortic bifurcation. It had the typical histologic appearance of paraganglionic tissue, but the chromaffin reaction was so questionable that the authors considered it to be negative. After two months' preservation in solution of formaldehyde, the tumor was tested for epinephrine; the Vulpian reaction was positive, but the iodine and mercury sublimate reactions were negative. A positive result was obtained when the extract was tested on the isolated heart and eye of a frog. This they believed to be good evidence that the tumor was definitely a chromaffinoma, and they stated further that from their findings it was evident that a positive chromaffin reaction was not necessary so long as the presence of epinephrine could be proved.

The tumors of the Zuckerkandl bodies in the case reported in this paper were equally interesting because they displayed the two most common characteristics of paraganglionic tissue: a typical histologic appearance, and a positive chromaffin reaction. However, this is the only instance in which this type of tumor has produced a positive chromaffin reaction and, also, has been definitely analyzed for epinephrine and found to contain none.

19. Nordmann, Martin, and Lebküchner, Eberhard: *Virchows Arch. f. path. Anat.* **280**:152, 1931.

Taking all these results into consideration, the question arises as to the relation of epinephrine to chromaffinity.

Biedl<sup>7</sup> and Wiesel,<sup>20</sup> at the beginning of this century, stated that extracts of pure chromaffin tissue, when injected into animals, raised the blood pressure. This they believed to be due to the action of epinephrine. Further, they said that if the tissue was exhausted of its secretion by overstimulation of the splanchnic nerves, it no longer raised the blood pressure or exhibited the chromaffin reaction. Therefore they concluded that epinephrine was the substance stained by the chromium salt. A number of investigators (Kohn,<sup>21</sup> Suzuki and Herde,<sup>3</sup> Rabin,<sup>22</sup> Eisenberg and Wallerstein,<sup>23</sup> Maximow,<sup>24</sup> Gérard, Cordier and Lison,<sup>25</sup> Stoerk and von Haberer,<sup>26</sup> Lucien and Parsiot<sup>27</sup>) have stated that the chromium salt is reduced by epinephrine and deposited as an insoluble compound in cells containing epinephrine. Recently (1932), Rogoff<sup>28</sup> summarized this association when he expressed the view that, in general, the chromaffinity of a tissue appeared to be roughly proportional to its content of epinephrine. However, he immediately qualified this statement by saying that in some cases the chromaffin reaction was weak or absent when the epinephrine content was high, and that there had never been any satisfactory demonstration of a reduction in chromaffinity after prolonged splanchnic stimulation during which time epinephrine was secreted in large amounts. This was borne out by the results in the case reported here in which the suprarenal medulla contained an abundance of epinephrine, but evidenced a weak chromaffin reaction. Added to this is the fact previously referred to, namely, that epinephrine never has been detected in the carotid body. Further, the histochemical findings in the reported cases of tumors of the Zuckerkandl body, including the case reported here, have never disclosed the simultaneous occurrence of the chromaffin reaction and epinephrine in the same tumor. Therefore, I am inclined to believe that the widely accepted theory of reaction between these two compounds is incorrect

20. Wiesel, J.: *Virchows Arch. f. path. Anat.* **176**:103, 1904; *Anat. Hefte* **19**: 481, 1902.

21. Kohn, Alfred: *Ergebn. d. Anat. u. Entwickelungsgesch.* **12**:253, 1902.

22. Rabin, C. B.: *Arch. Path.* **7**:228, 1929.

23. Eisenberg, A. A., and Wallerstein, H.: *Arch. Path.* **14**:818, 1932.

24. Maximow, A. A.: *Textbook of Histology*, Philadelphia, W. B. Saunders Company, 1930, p. 704.

25. Gérard, P.; Cordier, R., and Lison, L.: *Compt. rend. Soc. de biol.* **105**:876, 1930.

26. Stoerk, O., and von Haberer, H.: *Arch. f. mikr. Anat.* **72**:481, 1908.

27. Lucien, Maurice, and Parsiot, J. V.: *Glandes surrénales et organes chromaffines*, Paris, Société d'études scientifiques et médicales, 1913.

28. Rogoff, J. M.: The Suprarenal Bodies, in Cowdry, E. V.: *Special Cytology*, New York, Paul B. Hoeber, Inc., 1933, vol. 2, p. 871.

and that they are not related. If this is true, one is forced to accept one of two conclusions: Either the carotid and Zuckerkandl bodies and tumors of these bodies belong to the chromaffin system and the presence of epinephrine is not essential to the diagnosis, or it is essential, and none of the carotid bodies and only a few of the Zuckerkandl bodies may be so classified. A greater part of the existing evidence, therefore, favors the former conclusion, and this is further substantiated by the fact that in the case reported here the concurrence of tumors of both of these glands points toward a common ground of origin and development.

#### SUMMARY

A case has been described of concurrent tumors of the left carotid body and both Zuckerkandl bodies. The similarity of their histologic appearance to that of paraganglionic tissue associated with the positive chromaffin reaction of the tumors of the Zuckerkandl bodies classifies them in the paraganglionic system. However, they were analyzed for epinephrine and found to contain none. Four other cases of tumor of the Zuckerkandl body have been reviewed and in none have a positive chromaffin reaction and a positive test for epinephrine occurred simultaneously. Also, these two factors have been found to be inconsistent in examination of the suprarenal medulla, and the presence of epinephrine has never been demonstrated in the carotid body. Therefore, I am inclined to believe that there is no definite association between the chromaffin reaction and epinephrine, and the presence of epinephrine is not essential in the classification of a tissue as a part of the paraganglionic system.

## RHINO-ENCEPHALOCELE

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AND

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BROOKLYN

The following is a report of an unusual form of encephalocele in which the extracranial brain tissue apparently represents a developmental anomaly of the rhinencephalon. The resemblance of this lesion to the so-called nasal glioma will be pointed out. The case also presents certain of the features of tuberous sclerosis.

### REPORT OF A CASE

*History.*—H. V., a new-born female infant, was admitted to the service of Dr. Cameron Duncan on Dec. 25, 1931. The baby had been born at home and was brought to the hospital because of a tumor mass attached to the front part of the head.

She was the third child of normal parents. The other two children were living and showed no congenital abnormalities. No neuropathic heredity could be established from the history. The infant weighed 3,030 Gm. She appeared to be normally developed except for a tense, lobulated, bluish mass attached by a broad pedicle in the midline of the forehead at the base of the nose (fig. 1). The anterior aspect of the mass consisted of three domelike projections covered with thin bluish skin. Over the remainder of the mass and over the pedicle the skin covering was much thicker. By pressure the tumor could be partially reduced, only to refill immediately when the baby cried. The pedicle was firm, and no cranial opening could be palpated.

Examination with roentgen ray revealed a circular bony defect approximately 2 cm. in diameter in the midline just between the medial ends of the superciliary ridges. Otherwise the cranial bones appeared normal.

For four days the infant took its formula well. On the fifth day the temperature rose to 102 F.; the respirations became rapid, and physical signs of bronchopneumonia appeared. The clinical course was downhill until death on the seventh day. The clinical diagnosis was meningo-encephalocele and terminal bronchopneumonia.

*Autopsy.*—At autopsy the diagnosis of bronchopneumonia was confirmed, and a congenital defect of the interventricular septum of the heart was also found. Dr. W. W. Hala permitted us to use the encephalocele.

The tumor protruding from the frontal region of the head presented a different appearance from that noted during life. It was about one third as large; the lobulations were less definite; its skin covering was wrinkled, and its remaining contents could no longer be reduced. The mass was definitely pedunculated, being attached by a short thick pedicle in the midline of the forehead between the supra-orbital ridges.

A cutaneous incision was made about the base of the pedicle, disclosing a smooth-rimmed cranial defect. Thereupon the scalp was reflected by a midline incision and the skull opened by separating the cranial bones at the suture lines. On exposing the cerebral hemispheres a stalk of tissue was found connecting the inferior aspect of the frontal lobes of the brain with the extracranial mass. The dura, which passed through the bony defect, was incised to permit removal of the brain, the tumor and the connecting pedicle en masse.

Following the removal of the brain the dural covering of the anterior fossa was found to be smooth with no prominence in the region of the crista galli, no olfactory grooves and no perforations for the passage of the olfactory filaments. After the dura had been stripped away the cribriform plate of the ethmoid bone presented a smooth cartilaginous surface. Neither the intracranial cavity nor the region of the cranial defect communicated with the nasal cavities.



Fig. 1.—The infant shortly after birth. The tense lobulated mass is seen attached to the base of the nose.

After fixation in solution of formaldehyde the tumor measured 3.5 by 4 by 4 cm. It was firm and covered with thin wrinkled skin. It was attached to both frontal lobes of the brain by a broad pedicle.

As viewed from the dorsal aspect the cerebral hemispheres appeared grossly normal. The ventral aspect of the frontal region presented an unusual picture, a striking feature being the total absence of the olfactory bulbs and tracts (fig. 2). In their place there was a relatively broad elevated ridge made up of several irregular convolutions which coursed forward into the pedicle of the tumor. This area was covered with thickened pia-arachnoid enclosing large branches of the anterior cerebral arteries which also passed into the tumor. The remainder of the cerebral hemispheres, the brain stem and the cerebellar hemispheres, from this aspect, appeared anatomically normal except for a slight thickening of the arachnoid.

The membranes and the blood vessels were stripped from the frontal lobes and the stalk of the tumor. The arachnoid was found to continue into the tumor proper; in fact, a large nodule of the tumor lay external to this layer (fig. 2). A continuation of the median longitudinal fissure divided the pedicle into two unequal portions, the left portion, consisting of a single convolution, passing into the tumor as a smooth bulbous swelling which somewhat resembled an olfactory bulb. The right portion was formed by several convolutions, the most mesial of which resembled the one on the left side whereas those more laterally placed appeared as irregular prolongations of the orbital gyri. The latter convolutions were unusually small and numerous, becoming progressively smaller toward the

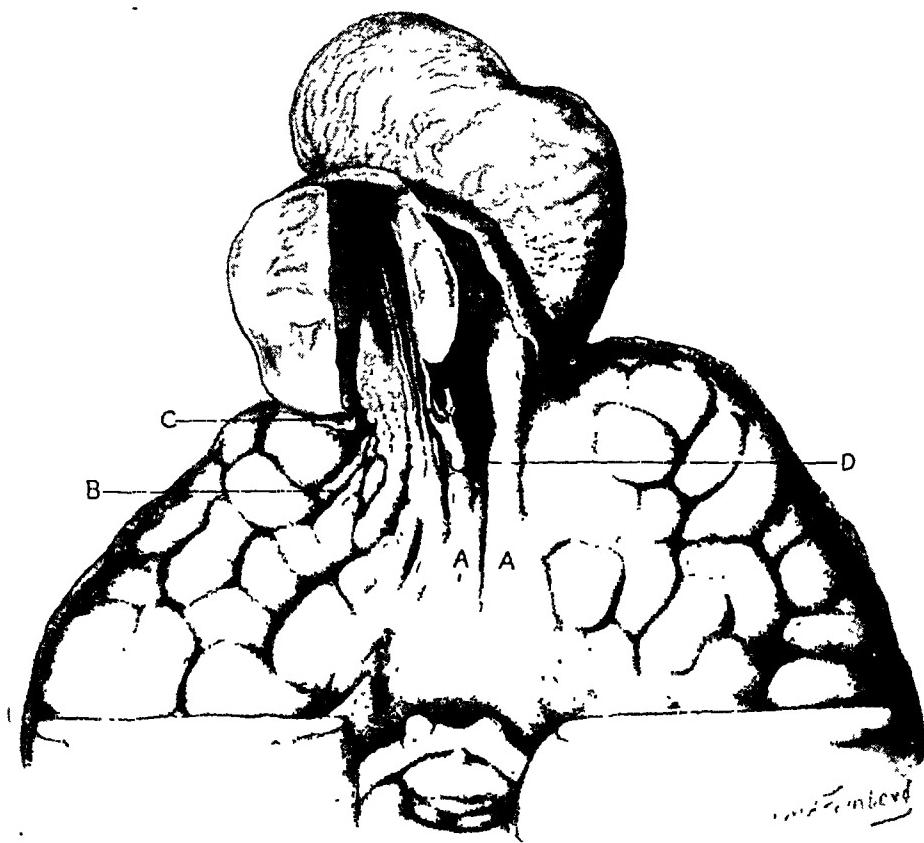


Fig. 2.—Anterior portion of the brain and tumor. The meninges have been stripped away to show: *A*, the absence of olfactory bulbs and tracts; *B*, an area of microgyria in the right orbital gyri and tumor pedicle; *C*, the arachnoid entering the tumor internal to portions of the tumor tissue, and *D*, the median longitudinal fissure.

base of the tumor where they appeared as small closely set nodules. The olfactory striae could not be identified. In this position there was a broad smooth surface bounded laterally by the anterior perforated spaces.

A sagittal section was made through the midline of the brain stem, passing as nearly as possible through the center of the tumor and its pedicle. In the pedicle this section passed somewhat to the right of the median longitudinal fissure, exposing a cavity which communicated with spaces within the tumor (fig. 3).

The structure of the tumor and its pedicle and their relationship to the brain proper can perhaps be described best on the basis of a combined gross and microscopic study, since our original interpretations, based on purely gross anatomic observations, had to be materially revised in the light of subsequent histologic revelations.

The tumor consisted essentially of two cystlike out-pouchings of brain tissue each having a central cavity which communicated, by a separate passage through the pedicle, with the anterior horns of the left and right lateral ventricles, respectively. Each of these pouches was covered with a hyperplastic layer of pia-arachnoid continuous with that of the brain proper. Fragments of dense connective tissue, apparently dura, formed an incomplete investment of both pouches and partially separated them from each other (fig. 4).

From the outer (pial) surfaces of the pouches numerous polypoid outgrowths projected into the pia-arachnoid, and even, through defects in the meningeal coverings, into the subcutaneous layer of the skin. The larger of these polypoid

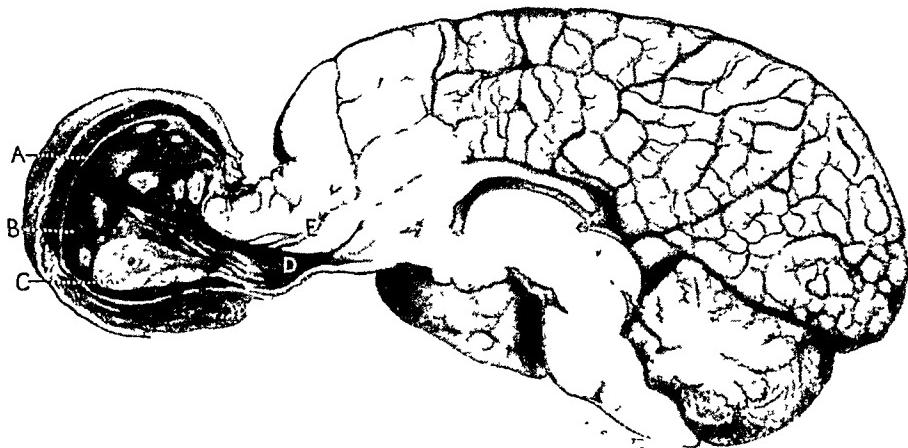


Fig. 3.—Mesial surface of the right hemisphere and corresponding portions of the tumor and pedicle; *A*, pouch of brain tissue; *B*, ependymal-lined cavity; *C*, polypoid structures protruding from the inner surface of the pouch; *D*, canal in the pedicle with its roof formed by *E*, convolutions of the frontal lobe.

structures measured from 0.5 to 1 cm. in average diameter while the smaller ones were of pinhead size or smaller, many being visible only microscopically.

Similar but generally larger polypoid structures projected from the inner (ventricular) surfaces of the pouches, filling their central cavities almost completely.

The portion of the tumor derived from the right hemisphere comprised about three fourths of the entire mass. The polypoid structures in its cavity were firm, of a tan color due to staining with hemoglobin, and for the most part large and smooth-surfaced. Many of them were branched. They had their origin near the neck of the pouch or arose by long narrow stalks from the wall of the cavity within the right portion of the pedicle.

The portion derived from the left hemisphere formed a flattened caplike mass with a small collapsed central cavity. The polypoid growths projecting into this cavity were much fewer and smaller than those found in the right portion of

the tumor. The outgrowths from the pial surface, however, were numerous, and the larger of these measured from 1 to 1.5 cm. in diameter.

The canal in the right portion of the tumor pedicle was relatively large. Its lining inferiorly and laterally was made up of parallel longitudinal folds of brain tissue which streamed forward into the body of the tumor and gave origin to

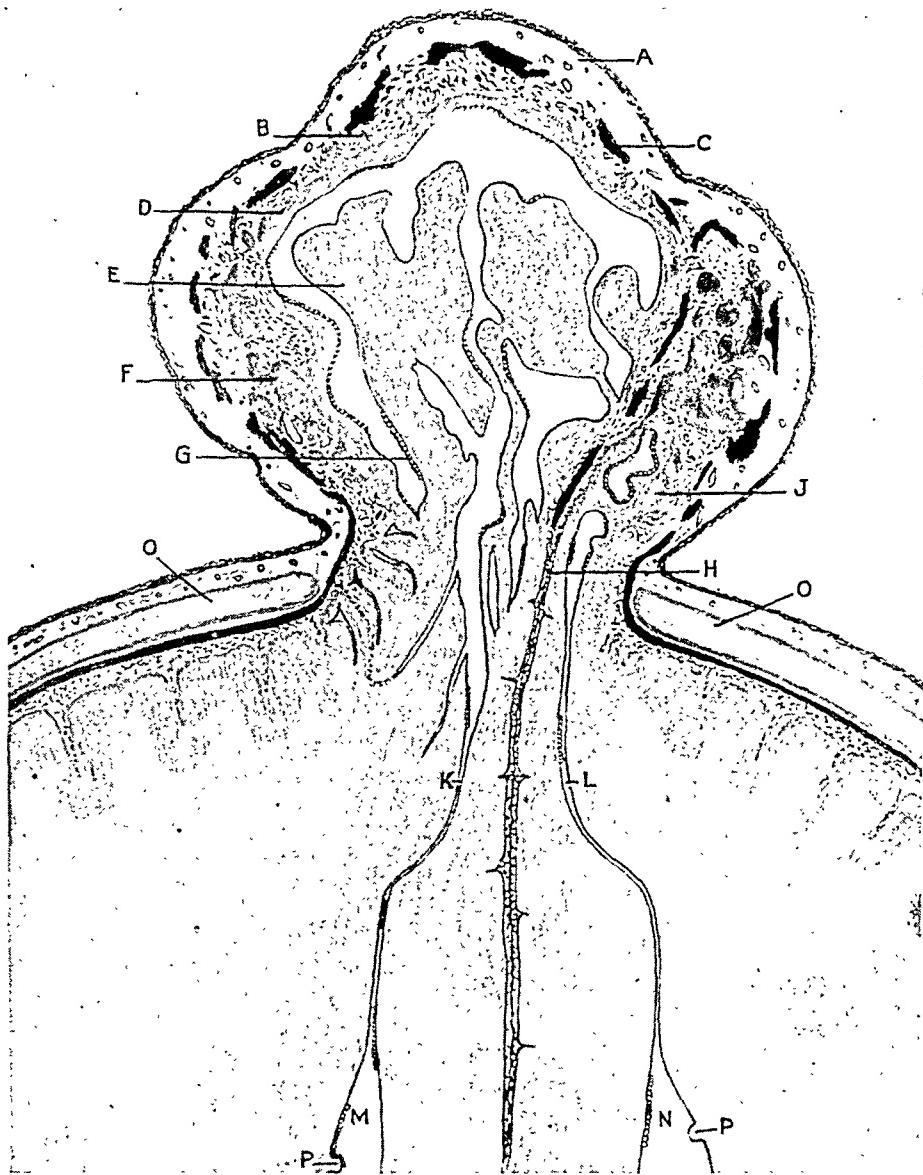


Fig. 4.—Diagrammatic representation, from below, of the encephalocele and its relationship to the cerebral ventricles: *A*, skin covering; *B*, subarachnoid space; *C*, fragments of dura; *D*, pouch of brain tissue derived from the right hemisphere; *E*, polypoid nodules projecting into its cavity and *F*, into the subarachnoid space; *G*, remnants of an ependymal lining; *H*, the median longitudinal fissure; *J*, the left pouch with its cavity and polypoid outgrowths; *K* and *L*, canals through the pedicle leading to *M* and *N*, right and left lateral ventricles; *O*, cranium with the nasofrontal defect, *P*, the subependymal nodules in the cerebral ventricles.

many of the polypoid structures noted. Superiorly the cavity was bounded by convolutions of the frontal lobe devoid of meningeal coverings and drawn forward and flattened so that they terminated in sharp thin edges. Among these convolutions communications could be traced between the ependymal-lined cavity and the subarachnoid space. As the canal traversed the frontal lobe of the brain it narrowed abruptly, curving outward and upward to join the anterior horn of the right lateral ventricle.

A much smaller canal was found in the left portion of the tumor pedicle. It could be traced backward to the anterior horn of the left ventricle.

The mesial aspect of the hemispheres presented various other abnormalities. The anterior portion of the corpus callosum was poorly defined, and the parolfactory area was not recognizable as such. The marginal and callosal gyri appeared drawn forward toward the base of the tumor. Indeed, several atypical convolutions of these gyri entered into the formation of the tumor pedicle and formed the superior wall of the canal in its right portion as noted. The remainder of the brain from this aspect was essentially normal.

Multiple sections in the coronal plane revealed numerous small nodular protrusions on the ependymal surfaces of all ventricles. These were most numerous in the fourth ventricle and in the occipital and temporal horns of the lateral ventricles. They varied from the size of a pinpoint to a diameter of 3 mm. Most of them bulged only slightly into the ventricles while several were pedunculated. They were slightly paler and somewhat firmer than the surrounding brain tissue. A row of small nodules arose from the under surface of the quadrigeminal body, protruding into the aqueduct of Sylvius but causing no apparent obstruction.

Several of the nodules became evident only in stained preparations since they did not protrude into the ventricles and were not distinguishable grossly on the basis of their color and consistency.

We estimated that there were between thirty and forty of these "subependymal" nodules exclusive of similar structures in the extracranial mass and its pedicle.

No discrete nodules or areas of sclerosis could be found in the cortical or subcortical zones of the hemispheres except where they entered into the formation of the encephalocele and its pedicle.

Microscopic study of the right portion of the extracranial mass showed a thin-walled pouch composed of sclerotic brain tissue in which there was a vague differentiation into cortical and medullary zones. The central cavity contained some fairly fresh blood, and its lining surface was superficially eroded so that it appeared rather ragged. In some areas, however, remnants of an ependymal lining could be recognized. The polypoid and nodular protrusions into the cavity were of the same general structure as the wall of the pouch. The outgrowths from the pial surfaces were numerous, and many of them were small. They lay in a richly vascular, thickened pia-arachnoidal layer. Some of them were attached by narrow stalks while others were widely separated from the wall of the pouch and were devoid of any direct attachment to it (fig. 5). (The latter observation was made through a study of drawings from serial sections, made by means of a camera lucida.) In some areas an intermingling of these processes with fibrotic pia-arachnoid suggested an invasive tendency on the part of the tumor tissue.

Fibrillary astrocytes predominated both in the wall of the pouch and in the structures protruding from its surfaces (fig. 6 C). Microglial cells were abundant along the eroded surfaces of the tumor nodules and adjacent to scattered small hemorrhages (figs. 6 A and B). Oligodendroglial cells were also numerous. Many cells could not be definitely identified. Neurons were found in moderate

numbers and showed various forms of degeneration (fig. 6 D). In some areas masses of myelinated fibers extended like a fringe from the surfaces of the nodules, and occasional long myelinated fibers could be traced through their substance. The structure of some of the nodular growths suggested central neurinoma.<sup>2</sup>

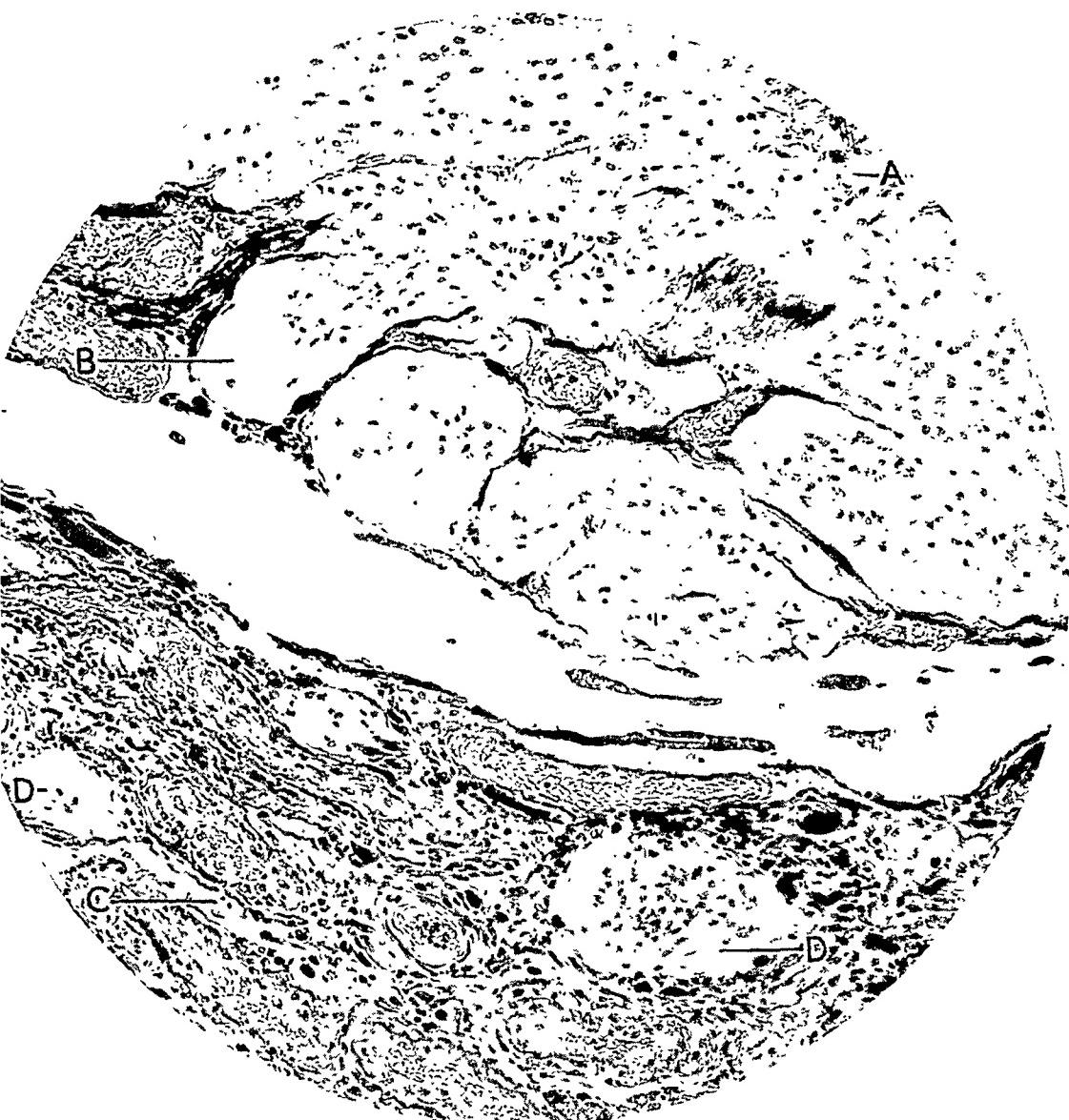


Fig. 5.—*A*, the wall of one of the pouches with *B*, nodular protrusions into *C*, the hyperplastic pia-arachnoid. At *D* are isolated nodules of the same character (hematoxylin and eosin).

The left portion of the extracranial mass was essentially the same as the right in its microscopic details.

2. This resemblance was pointed out by Dr. Joseph H. Globus with whom we discussed the case and to whom we are indebted for the name "rhino-encephalocele."

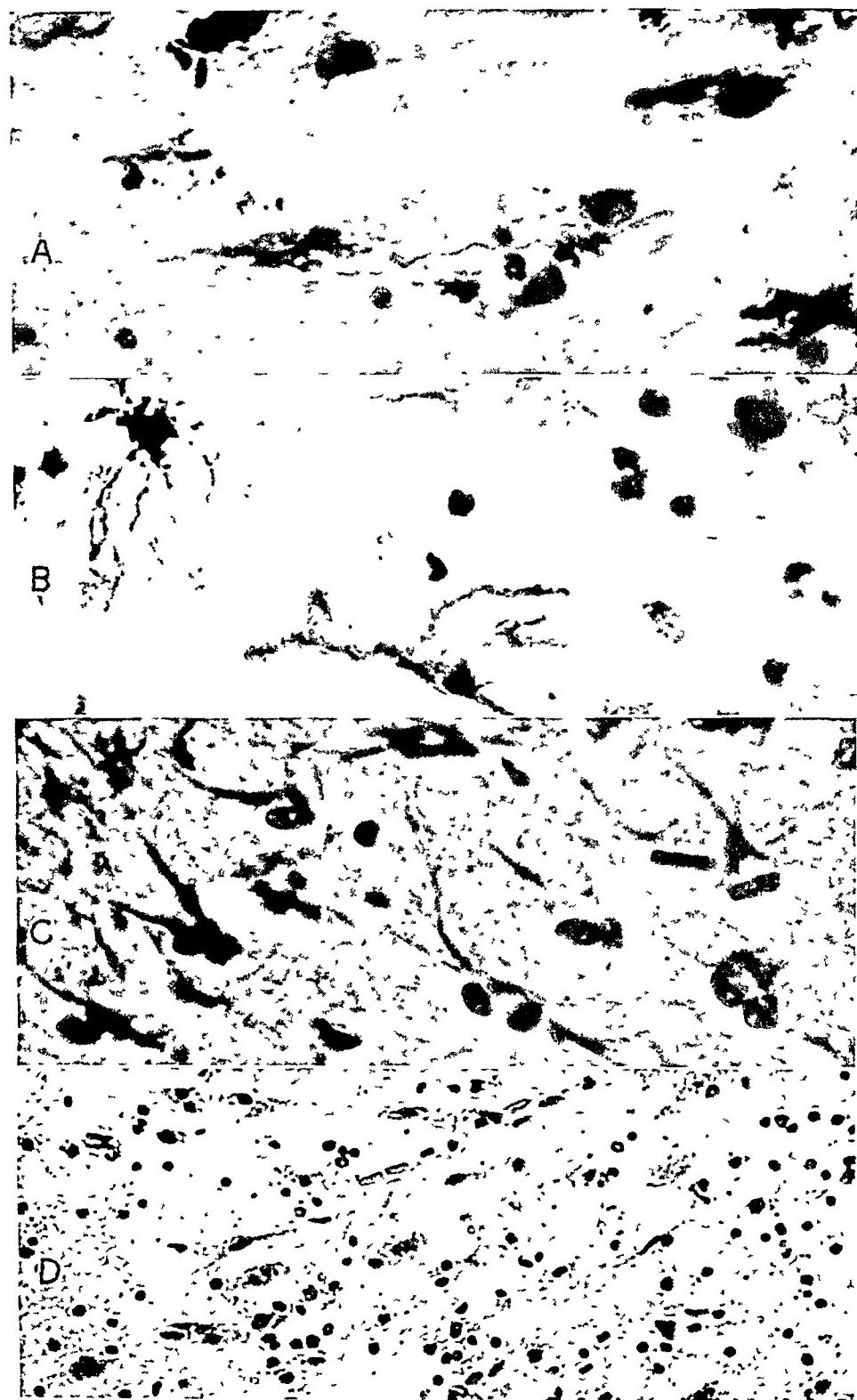


Fig. 6.—Cellular components of the nodules of the tumor, *A* and *B*, microglia (Penfield); *C*, fibrillary astrocytes (Loughlin method), and *D*, neurons (Nissl).

In the pedicle of the tumor an interesting feature was the extreme thinness of the walls of the ventricular cavities. In this region also fragments of ependyma persisted. In the distal portion of the pedicle the brain tissue was markedly sclerotic while more centrally the gliosis was rather closely confined to a zone about the ventricular cavities and merged by a gradual transition with relatively normal surrounding brain tissue.

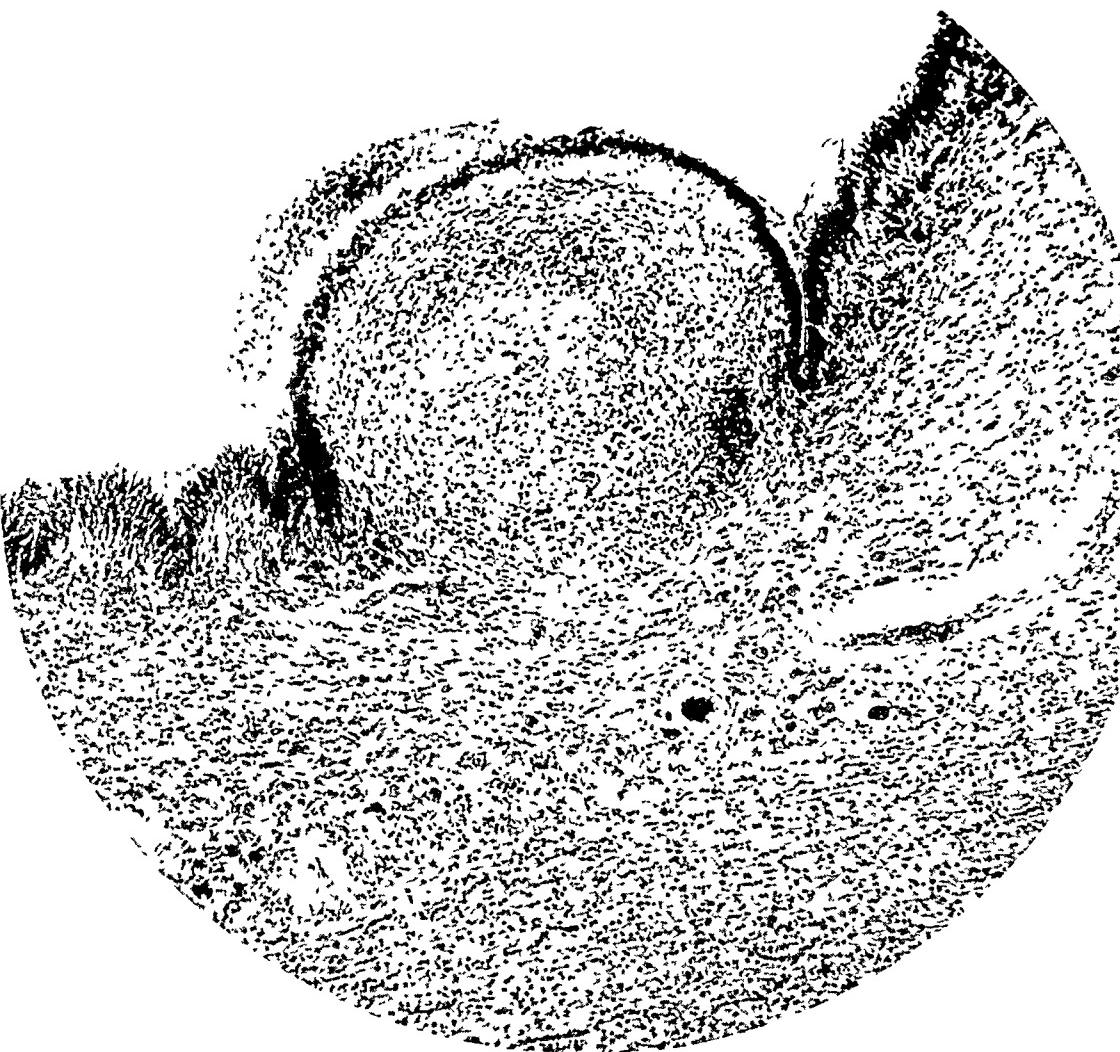


Fig. 7.—One of the smaller of the subependymal nodules protruding into the cerebral ventricles.

The nodules protruding into the ventricular cavities of the brain proper consisted of small discrete cellular areas sharply demarcated from the surrounding white matter. Some were covered on their ventricular surfaces by hyperplastic and irregular ependyma (fig. 7), while others showed eroded surfaces. The nuclei in the nodules appeared round or oval, and no cell bodies or processes could be

demonstrated by the ordinary staining procedures. By means of an apparently new staining method<sup>3</sup> we were able to demonstrate the glial nature of many of the cells. They were predominantly fibrillary astrocytes with smaller numbers of protoplasmic astrocytes, oligodendroglial and microglial cells. No neurons could be recognized. In the adjacent white matter was a zone of compressed nerve fibers with numerous microglial cells.

The anterior horns of both lateral ventricles were collapsed and partially obliterated by gliosis. Many sections from grossly normal portions of the brain showed retarded development in the cortical architecture. Only a slight degree of myelinization could be demonstrated, perhaps owing in part to unfavorable fixation. Frequently neurons were seen in the subcortical white matter. No mitral cells were found either in the neighborhood of their usual location or in the extracranial brain tissue. Sections through the hippocampal gyri showed a suggestion of the characteristic grouping of cells into islets, but the cells appeared fewer and less regularly arranged than usual. The dentate gyri also resembled the normal but were poor in nuclear elements and somewhat atypical in architecture. The cerebellum showed a broad external granular layer.

#### COMMENT

The case presents as its outstanding features: (1) a meningoencephalocele in which the protruding brain tissue consisted of sclerotic out-pouchings from the inferior portions of the frontal regions of both cerebral hemispheres, (2) an absence of recognizable olfactory lobes, tracts and striae, (3) an absence of perforations in the cribriform plate of the ethmoid and of the crista galli, (4) subependymal nodules scattered throughout the ventricular system, (5) histologically similar nodules protruding from the inner and outer surfaces of the extracranial cerebral out-pouchings, (6) a developmental anomaly in another organ, namely, a defective interventricular septum of the heart.

Grossly the extracranial mass is reminiscent of the rhinencephalon of the lower mammals. We believe that this structure represents an abnormally developed rhinencephalon. Its nodular and polypoid protrusions we interpret as the expression of a blastomatous character acquired by the extracranial brain tissue.

The stage of development of the embryo at which this disturbance must have originated may be roughly estimated as being at least prior to the third month when normally the rhinencephalon is found well differentiated into olfactory bulbs and tracts and the cartilaginous cribriform plate of the ethmoid has already formed around the olfactory nerve fibers. By this time the crista galli is normally represented by a cartilaginous ridge.

The subependymal nodules bear a close resemblance to those found in tuberous sclerosis except for the absence of the characteristic "giant cells." The presence of additional developmental anomalies is also sug-

3. This method which combines features of the Cajal and Bielschowsky procedures will be reported elsewhere by its originator, Dr. E. H. Loughlin.

gestive of tuberous sclerosis. On the other hand, no definite cortical sclerotic nodules were found, and the heterotopic ganglion cells in the subcortical zone were not of the abnormal types described in that condition. We believe, therefore, that this case cannot be considered an example of tuberous sclerosis although many features indicate a relationship to that disease.

We have gathered from the literature reports of about fifty cases in which tissue, more or less resembling brain tissue, has been found subcutaneously at the base of the nose, in the nasal or nasopharyngeal cavity or in both locations. Most of these were evidently instances of congenital malformations and have been variously designated as sincipital or basal hernias of the brain, encephaloceles, encephalomas, gliomas or fibrogliomas.

From an anatomic standpoint the lesions, which are most frequently referred to as hernias of the brain or encephaloceles, may be divided into sincipital and basal types. According to von Meyer<sup>4</sup> there are three subdivisions of the former:

1. Encephalocele nasofrontalis, in which the pedicle passes through a cranial defect located at the junction of the nasal and frontal bones. In such cases the tumor is externally visible in the midline at the base of the nose.

2. Encephalocele naso-ethmoidalis, in which the bony defect is between the frontal, nasal and ethmoid bones, and the tumor appears externally in the neighborhood of the junction of the bony and cartilaginous portions of the nose.

3. Encephalocele naso-orbitalis. In this instance the cranial defect lies between the frontal, nasal and lacrimal bones. The tumor enters the orbit, appearing at or near the inner canthus of the eye.

As Fenger<sup>5</sup> has noted, the naso-ethmoidal and naso-orbital varieties are not easily distinguished from each other since they leave the cranium at about the same place.

Basal encephaloceles, sometimes considered a subdivision of sincipital encephaloceles, are distinguished from the foregoing types by forming tumors not usually visible on the face. Of these Heinecke<sup>6</sup> described three forms:

1. Encephalocele spheno-pharyngealis, which leaves the cranium between the body of the sphenoid and the ethmoid bone and extends into the nasal or nasopharyngeal cavities or even into the mouth.

2. Encephalocele spheno-orbitalis, which leaves the cranium through the superior orbital fissure to enter the posterior portion of the orbit.

4. von Meyer, Edward: Virchows Arch. f. path. Anat. **120**:309, 1890.

5. Fenger, Christian: Am. J. M. Sc. **109**:1, 1895.

6. Heinecke, W., quoted by Fenger.<sup>5</sup>

3. Encephalocele spheno-maxillaris which passes through both the superior and the inferior orbital fissures into the spheno-maxillary fossa. "The tumor can be felt in the mouth on the medial side of the ascending ramus of the inferior maxilla, and is visible on the outside of the face, on the cheek below the zygoma, in the same place where the retro-maxillary branches of retro-nasal fibroids present" (Fenger<sup>5</sup>).

It seems possible that the rhinencephalon may be the portion of embryonal brain particularly concerned in the formation of encephaloceles in any of these locations. Our review of reported cases, however, has not included the many instances of orbital encephalocele, since these seem less likely to be genetically similar to the case here reported.

We recognize that in the large group of cases which we have reviewed the structural characteristics of the extracranial tumors are widely varied. In 1890 Berger<sup>7</sup> observed:

The structure of the nerve tissue which enters into the formation of certain encephaloceles departs notably from the type of the normal brain parts from which the tumors are derived. Because of this circumstance and the modifications in their meningeal envelops they ought to be considered as true neoplastic products to which the name encephaloma should be given.

Von Meyer<sup>4</sup> reported as an instance of basal hernia of the brain the case of an infant with an intranasal polypoid growth composed of solid glial tissue. The tumor was attached to the brain by a stalk which passed through an opening in the ethmoid bone. Fenger<sup>5</sup> reported the successful extirpation of a similar intranasal growth which he likewise called a basal hernia. In fact, most of the older records of cases of this general nature appear under the designation of hernias of the brain, although in many instances the herniated tissue showed definite blastomatous characteristics.

Extranasal or intranasal growths of this character have been called gliomas by Schmidt,<sup>8</sup> Clark<sup>9</sup> and Sussenguth,<sup>10</sup> and fibrogliomas by Rocher and Anglade,<sup>11</sup> while Terplan and Rudofsky<sup>12</sup> pointed out the resemblance of an intranasal growth to neurinoma.

Among the cases reported as encephalocele are many in which there were marked blastomatous characters. The cases of Schötz,<sup>13</sup> Guthrie and Dott,<sup>14</sup> Natanson<sup>15</sup> and Malek<sup>16</sup> fall into this group.

7. Berger, Paul: *Rev. de chir.*, Paris **10**:269, 1890.

8. Schmidt, Martin B.: *Virchows Arch. f. path. Anat.* **162**:340, 1900.

9. Clark, Payson: *Am. J. M. Sc.* **129**:769, 1905.

10. Sussenguth, L.: *Virchows Arch. f. path. Anat.* **195**:537, 1909.

11. Rocher, H. L., and Anglade: *Rev. de chir.*, Paris **62**:147, 1924.

12. Terplan, K., and Rudofsky, F.: *Ztschr. f. Hals-, Nasen- u. Ohrenh.* **14**:260, 1926.

13. Schötz, W.: *Ztschr. f. Hals-, Nasen- u. Ohrenh.* **58**:137, 1909.

14. Guthrie, D., and Dott, N.: *J. Laryng. & Otol.* **42**:733, 1927.

15. Natanson, Leo: *Arch. f. Ohren-, Nasen- u. Kehlkopfh.* **135**:103, 1933.

16. Malek, S. A.: *J. Anat.* **66**:264, 1932.

Berblinger,<sup>17</sup> reporting a case of glioma at the base of the nose, suggested the possibility of a developmental relationship to the olfactory bulb. Terplan and Rudofsky<sup>12</sup> accepted Berblinger's theory so far as the structure of the tumor in their case permits of this possibility. In both instances, however, no confirmation of this idea was possible.

An isolated instance in which there was definite evidence of a relationship between the olfactory bulbs and "an encephalocele in the nasal region" has been reported by Malek.<sup>16</sup> The case was that of a new-born infant with a large pear-shaped tumor situated in the mid-line of the face, extending from the middle of the forehead to the upper lip. The encephalocele, evidently belonging to the naso-ethmoidal group, was covered by dura and skin, contained ependyma-lined cavities and was attached to the brain by a pedicle passing through a cartilaginous tube. Within the cranial cavity the pedicle divided into left and right portions, each of which was attached to the inner margin of the corresponding olfactory bulb. The extracranial mass was described as a glioma. The author concluded: "The tumor is of cerebral origin and appears to be a protrusion from that part of the neural tube which gave rise to the olfactory bulbs. This is indicated by its connection with the latter and by the ependymal-lined cavities contained in it."

That other reported encephaloceles in this region may have been derived from the rhinencephalon seems highly probable, but we have been unable to find any other case presenting sufficient evidence to establish this relationship. On the other hand, it seems even more probable that not all such tumors were "rhino-encephaloceles." But the evidence in support of this statement is also unsatisfactory, for in only a few instances has the condition of the olfactory bulbs been recorded. Even in the reports of encephaloceles leaving the cranium through a defective lamina cribrosa the olfactory bulbs almost invariably are not mentioned.

We have recently reexamined the tissue from a case reported by one of us (Browder<sup>18</sup>) and found a striking similarity to the tumor in the case reported here. A brief summary of this case will serve to emphasize this resemblance.

L. V., an infant, aged 11 months, showed a nodular mass "about the size of an English walnut" at the base of the nose in the midline. A previous attempt at removal of the mass had been abandoned because of excessive hemorrhage. Examination with roentgen ray showed "a circular bony defect in the region of the glabella." At operation the mass was freed from below upward until a stalk was identified which led through the small bony opening in the skull. The opening was enlarged; the dura was incised, and the stalk was seen to divide into two portions, the larger part being attached to the right frontal lobe, the lesser to the

17. Berblinger, quoted by Terplan and Rudofsky.<sup>12</sup>

18. Browder, J.: Ann. Otol., Rhin. & Laryng. 38:395, 1929.

left. Both attachments were divided. There was no evident communication with the ventricles. The postoperative course was good for the first twenty-four hours; then bronchopneumonia developed, and the child died. There was no evidence of meningitis. Permission for an autopsy was refused. The excised tissue contained a soft, grayish-white central mass from which smaller nodules of similar consistency extended into the surrounding vascular connective tissue. The free surface was covered with normal skin. The microscopic sections showed a structure almost identical with portions of the tumor in H. V. (fig. 5).

In this case as in many others in the literature the possible derivation of the tumor from the rhinencephalon remains a matter of speculation.

#### CONCLUSION

The case of an infant with an unusual type of nasofrontal encephalocele which we interpret as a developmental anomaly of the rhinencephalon is recorded. The blastomatous features of the encephalocele are regarded as secondary characteristics such as have been indicated in previous reports by the names "nasal glioma," "encephaloma" and "fibroangioma." We have used the designation "rhino-encephalocele" to indicate the origin and essential nature of the lesion.

The presence of certain of the features of tuberous sclerosis suggests a genetic relationship between the two processes.

# STUDIES IN ATHEROSCLEROSIS: CHEMICAL, EXPERIMENTAL AND MORPHOLOGIC

III AND IV. RÔLES OF CHOLESTEROL METABOLISM, BLOOD PRESSURE  
AND STRUCTURE OF THE AORTA; THE FAT ANGLE OF THE  
AORTA (F.A.A.), AND THE INFILTRATION-EXPRESSION  
THEORY OF LIPOID DEPOSIT

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## III. RELATIONSHIP OF PHYSICAL CHEMICAL CHANGES IN THE AORTA TO INCLINATION TO ATHERO- SCLEROSIS (F.A.A.)

The problem of determining abnormal states in the aortic wall is difficult. The difficulty becomes more acute when one considers that the normal changes occurring in this vessel are not definitely established.

The two great factors that may alter the structure of the aorta are age and blood pressure. Endowment, naturally, forms the basis from which these components must be measured.

### A. THE RELATIONSHIP OF AGE AND THE INTERNAL ELASTIC MEM- BRANE TO THE LIPOID DEPOSIT IN THE AORTA

*Age.*—The important alterations of the aorta with age are given as changes in the interstitial substance and elastic lamellae which finally lead to a dilatation of the vessel.

The Interstitial Substance: The nature of the interstitial substance and the elastic fibers of the aorta has given a subject for much discussion. The connection between the elastic fibers and the interstitial substance is not well understood, but Benninghoff has shown that the interstitial substance appears before the elastic fibers, and that the latter, when developed, are contained in the former, thus forming one continuous sheet. The latter is impregnated with numerous channels for drainage.

With increasing age the amount of the interstitial substance is reported to be increased (Bjorling, Schultz, Ssolowjew,<sup>a, b</sup> Cellina). This probably means that the increase is only relative, as the elastic fibers decrease (Benninghoff). Although it was at first thought that this augmentation was the underlying factor in the development of atherosclerosis, it was later established that it was physiologic and was found in old persons without atherosclerotic changes of the aorta (Björling, Schultz, Ssolowjew,<sup>a, b</sup> Cellina). That this substance does alter with age as do all aging colloids (Wells), and that it then has an affinity for fat and calcium, are universally admitted (Aschoff,<sup>d</sup>

Wells, Okunoff). Whether this depends on the changed colloid state or whether it is related to the increased acidity (Schmidtman and Huttich) is unknown. That it is not the only factor involved in atherosclerosis has been clearly shown by Björling, Schultz, and others.

The Internal Elastic Membrane: The intima at birth is negligible but soon after develops rapidly into three layers: an inner fibrous layer, a middle fibro-elastic layer bounded distally by an external limiting membrane, and an outer fibromuscular layer terminated by the internal elastic membrane (Key-Aberg, Grundstein, Jores, Voigt, A. Aschoff).

According to A. Aschoff, this development reaches its peak at about 33 years (ascending stage of life). From then on there is a stationary period until 45 years, after which degenerative changes begin (descending stage of life).

The cause of this hypertrophic, hyperplastic development of the aortic intima and its relation to atherosclerosis have been greatly disputed points. Many authors believe that it is secondary to the irritation caused by the lipoid deposit (Anitschkow<sup>b, c</sup>), which occurs from birth onward.

Against this hypothesis are numerous facts. The lipoid deposits in youth, although to be found in the same sites as the atherosclerotic lesions (Zinzerling<sup>a</sup>), are not uniform and are most pronounced in the thoracic portion of the aorta. Yet, in a series of microscopic studies of human aortas, the development of the fibro-elastic layer in the abdominal portion was found to be much more pronounced than that in the thoracic portion. In the latter the fibro-elastic layer was about one-third to one-fourth as wide as in the former (figs. 3A and B).

The fat deposit was found to be in no way parallel to the proliferation of the internal elastic membrane. The fat deposit early in life was very slight, while the proliferation was extensive (Görög). In 11 of 28 cases Görög found that there was no lipoid in the internal elastic membrane under 20 years.

Experimentally in rabbits no proliferation of the elastic fibers was found after one hundred and twenty days of feeding cholesterol (0.5 Gm. in 10 cc. of linseed oil daily). Anitschkow<sup>c</sup> and Ssolowjew<sup>c</sup>, Stuckey, McMeans<sup>b</sup> and Wolkoff<sup>b</sup> reported elastic fiber increase after about six months of feeding, but, as Wolkoff<sup>b</sup> brought out, the animals in which these changes occurred were old ones.

According to Benninghoff, the structure of a vessel is dependent on the circulatory demands made on it and not on its endowment. Görög compared the increase in the volume of the blood with the size of the aorta from birth to adult life, and found that there was a great disproportion between the two. At birth the volume of blood is from 350 to 400 Gm., and in adult life it is from 5,000 to 6,000 Gm. The diameter

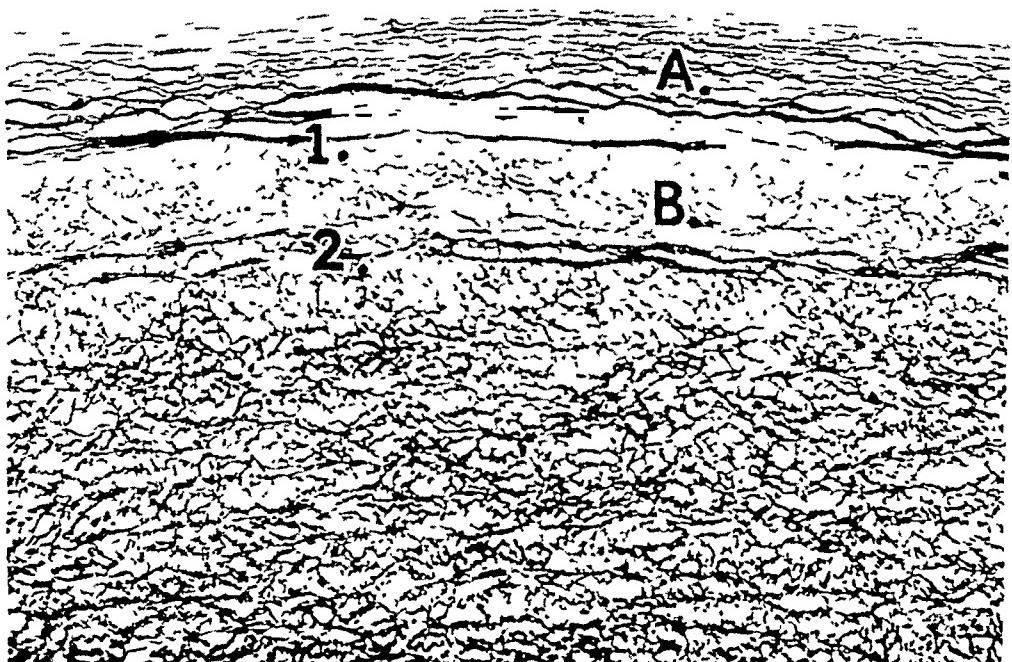
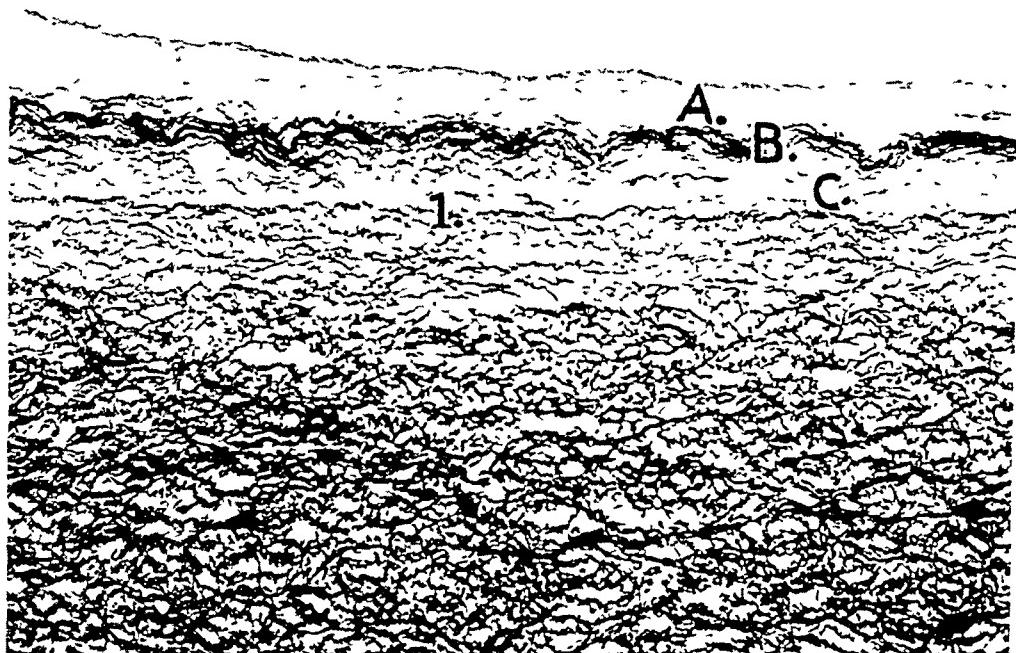


Fig. 3.—Aorta of a 35 year old normal woman: The upper half shows the ascending portion of the aorta, in which *A* indicates the fibrous layer, *B* the fibro-elastic layer, *C* the fibromuscular layer and *I* the internal elastic membrane. The lower half shows the abdominal portion in which *A* indicates the fibro-elastic layer, *B* the fibromuscular layer, *I* the external limiting membrane and *2* the internal elastic membrane. (Weigert's elastica stain;  $\times 100$ .)

of the aorta varies from 0.77 to 3.69—3.85 cm. (female and male, respectively), while the thickness varies from 1.17 to 1.71—1.84 mm. (male and female, respectively). He concluded that the size of the aorta, especially the thickness, cannot keep pace with the increase in blood volume.

The determination of the varying demands made on the aorta is best seen by its increase in size. Both the width and the thickness increase with age, the thickness increasing at the expense of the intima (Kani). Although the width of the aorta varies somewhat with the length of the body (Scheel), when a large group of cases is considered this difference is compensated for.

TABLE 22.—*The Measurements of the Aorta of the White and Colored Races from Birth to 30 Years of Age*

	White									Colored								
	Male			Female			Male			Female								
	Measurements, Mm.*			Measurements, Mm.			Measurements, Mm.			Measurements, Mm.								
	Age	Cases	1st	2nd	3rd	Cases	1st	2nd	3rd	Cases	1st	2nd	3rd	Cases	1st	2nd	3rd	
0-1	65	23.5	16.6	12.4	45	25.7	15.8	11.4	55	24.1	16.1	11.0	54	25.6	16.5	12.3		
2-5	43	37.0	22.6	16.8	31	35.6	21.7	15.9	47	37.0	24.1	16.7	42	36.0	22.2	15.9		
5-11	40	42.3	25.5	19.4	23	41.5	25.5	18.4	30	43.0	27.4	19.0	19	40.0	26.7	19.7		
11-15	23	50.4	31.6	25.1	13	50.2	29.0	21.8	11	46.4	31.4	22.3	15	45.5	28.8	19.4		
16-20	30	56.4	35.8	26.9	20	52.3	33.8	24.3	16	54.9	38.4	27.4	23	50.0	32.3	25.0		
21-30	41	58.0	38.2	30.3	29	55.2	35.9	28.1	31	61.5	39.5	29.9	53	55.7	36.0	28.2		
% increase	127	130	150			115	125	155		155	151	173		117	110	130		

\* The first measurement was above the aortic cusps, the second at the diaphragm and the third at the bifurcation at the aorta.

Table 22 presents the measurements of the aorta from birth to 1 year up to 30 years for both sexes and races. The three measurements of the aorta given were determined by a firm rule in the following locations: 1 cm. above the aortic cusps (first), at the diaphragm (second) and at the bifurcation of the aorta (third).

For both races and sexes there was practically no difference in the size of the aorta up to 11 years (also found by Rössle for both sexes of the white race). From then on the female aorta remained somewhat smaller in its second and third measurements, and from 16 years onward the first measurement also remained smaller. This is a clear demonstration of the effect of demand on the size of the aorta, as the young girl after 11 years is less active physically than the boy.

In comparing the increase in the ascending portion with that in the abdominal portion of the aorta there was usually about a 25 per cent increase of the latter over the former. This difference was probably due to the greater physiologic demands made on that part of the aorta and

may account for the difference in the development of the intima (Aschoff,<sup>f</sup> Jores, Benninghoff, Kani).

The thickness of the entire abdominal portion of the aorta also increases to a greater extent than that of the thoracic part. Kani showed that the ascending aorta increases in thickness from birth to 2 years up to 30 years, 49 and 29 per cent, respectively, for male and female. At the diaphragm this increase is 71 and 54 per cent, respectively, and at the bifurcation of the aorta 62 and 51 per cent, respectively.

What relationship there is between this fully developed intima and the lipoid deposition that follows has been much debated. Following the revival of the imbibition-infiltration theory of Virchow by Aschoff,<sup>e</sup> this concept has received recognition everywhere (Ribbert, Hueck, Jores, Anitschkow,<sup>a</sup> Petroff, Glasunow, Hackel, Lange<sup>a</sup>).

*One of the objections to this theory as it stands is that the lipoid in the serum as well as the arterial pressure is similar in the entire arterial tree including the arterioles (Lange<sup>c</sup>), yet the lipoid deposit is found mainly in the elastic type of arteries (aorta, pulmonary, coronary and carotid arteries), where the internal elastic membrane is poorly developed. Rarely is fat found in the muscular arteries, where the internal elastic membrane is well developed (Hesse, Lotzmann, Nordmeyer).*

Why this difference should exist has not been made clear, for if the internal elastic membrane acted as a barrier to the infiltrating lipoid from the blood plasma, one would expect lipoids to be deposited in the muscular arteries also.

A plausible solution of this problem was seen in the arteries of the cholesterol-fed rabbits which received 0.5 Gm. of cholesterol dissolved in linseed oil daily for one hundred and twenty days.

First, the normal histology of the rabbit's aorta must be considered (fig. 4 A). The intima consists of a layer of endothelium and a few fibrous and elastic fibers. There is no definite internal or external elastic membrane, and the media is rich in elastic fibers and, to a less extent, muscle fibers. The adventitia is insignificant. The peripheral vessels, as in man, are muscular and have a pronounced internal elastic membrane (fig. 4 B).

After feeding cholesterol, a deposit of lipoid readily occurred in the rabbit's aorta as well as in the pulmonary artery, but only rarely were the muscular arteries involved. In the aorta as in the pulmonary artery the lipoid deposit first occurred free beneath the endothelial lining, causing the intima to become wider. Later the deposit extended between the elastic fibers of the inner media. The fat deposit, however, instead of being found on the proximal aspects of the elastic fibers, was usually found on the distal aspects. In other words, although the cholesterol esters unquestionably filtered into the wall with the blood serum, there was probably an attempt to express the latter with contraction of the



Fig. 4.—*A*, normal rabbit's aorta. Note the insignificance of the intima, the internal and the external elastic membranes. (Weigert's elastica stain;  $\times 150$ .) *B*, muscular type of artery in the rabbit showing the well developed furrowed internal elastica membrane. (Weigert's elastica stain;  $\times 75$ .)

vessel. To further substantiate the possibility that a definite attempt was made by the elastic tissue to express the infiltrating lipoids, the peripheral vessels were examined. As has been stated, these vessels have a pronounced internal elastic membrane. Lipoid-staining material was found in the lumens of these vessels (fig. 5), but the intima only rarely showed lipoid deposit. In one such vessel a diffuse cholesterol deposit was found, whereas similar arteries in the vicinity were lipoid-free.

An examination of the internal elastic membrane of the latter vessel revealed that for some reason it had given way, and the fragments were displaced away from the lumen by the infiltrating cholesterol-laden

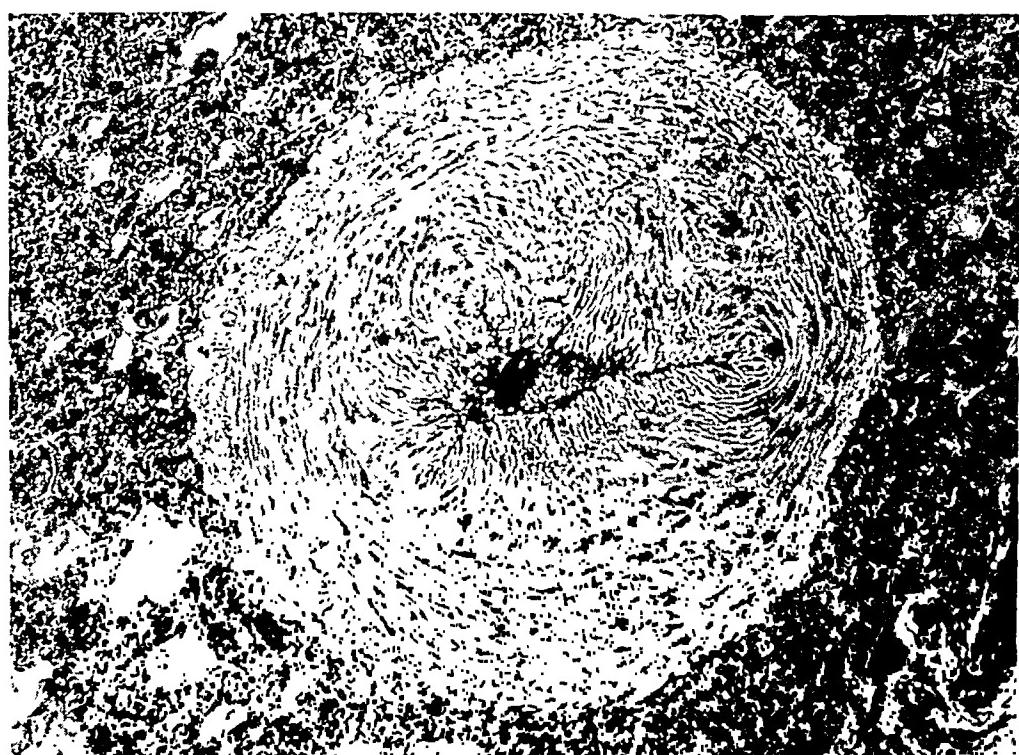


Fig. 5.—Muscular artery of rabbit. Note the fat in the lumen of the vessel and the absence of fat in the intima (dark-staining substance). (Sudan III;  $\times 75$ .)

serum (fig. 6 A). It seemed that after the internal elastic membrane had been destroyed, the infiltration of lipoids with the serum was unchecked and the expression of the same fruitless, accounting for the overwhelming diffuse deposit found there (fig. 6 B).

As one approached the arterioles, where the internal elastic membrane disappeared, lipoid deposit was again occasionally evident in the intima (liver, spleen, kidney, etc.).

From a physical standpoint the infiltration of cholesterol in the aorta is dependent on the cholesterol content of the serum, the blood pressure and the nature of the elastic membranes. In the rabbit's aorta, where

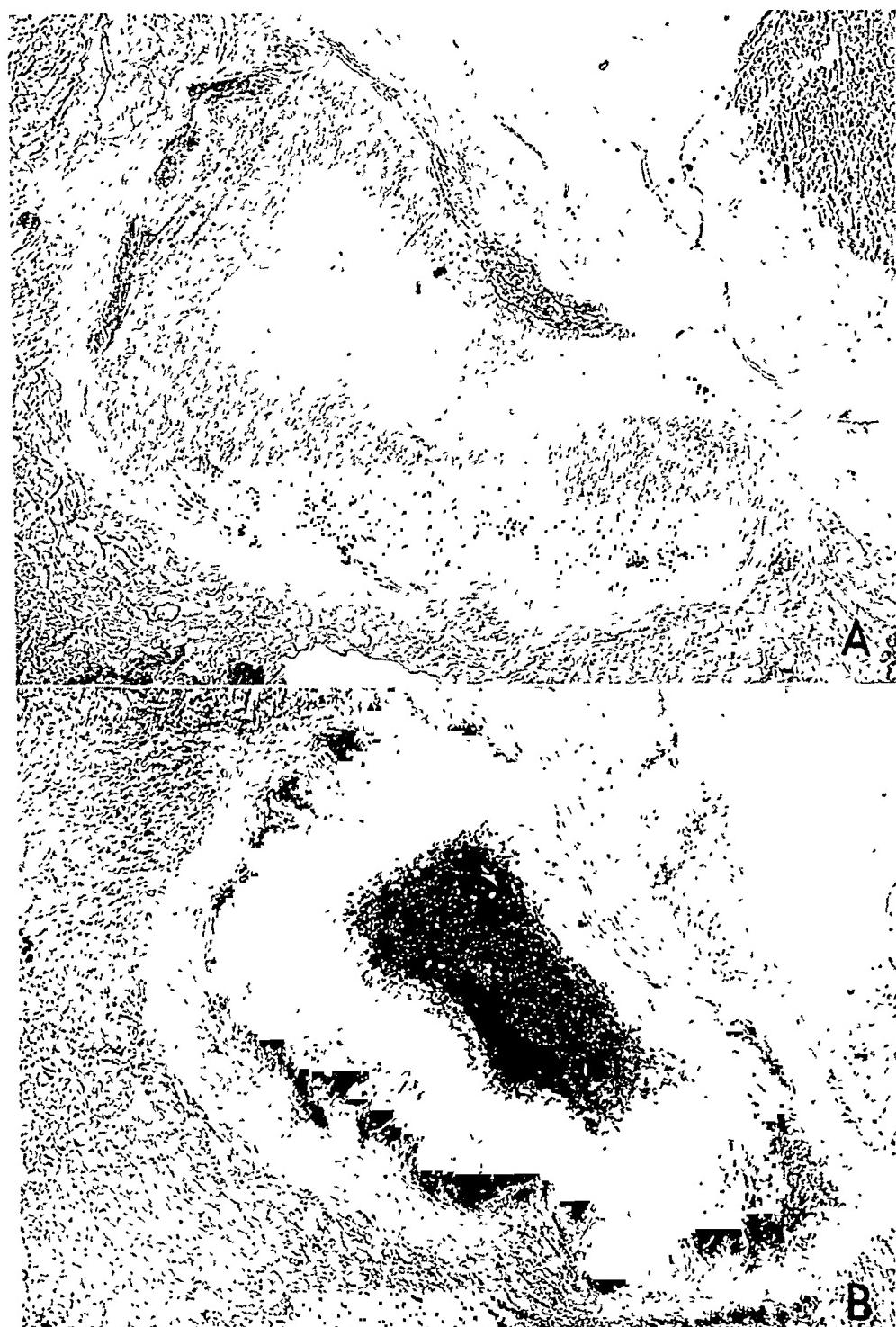


Fig. 6.—A, muscular artery of a rabbit showing fragments of the internal elastic membrane (dark-staining fibers). (Weigert's elastica stain;  $\times 75$ .) B, same artery showing marked fat deposit in the intima and the media (dark-staining substance). (Sudan III;  $\times 75$ .)

no definite elastic limiting membranes are present, the lipoid-containing serum during systole is pressed into the wall beneath the endothelium and between the elastic fibers. When cholesterol is present in increased quantities in the serum it may be deposited there. In the peripheral vessels the lipoids are arrested at the well developed internal elastic membranes.

The expression of the cholesterol-containing serum that occurs with the diastolic recoil of the vessel is dependent on the structure of the elastic limiting membranes, the latter's distance from the lumen of the vessel, and the irregularity of the contracting parts. In the aorta of the



Fig 7.—Early fat deposit in the abdominal portion of the aorta (dark-staining substance). Note that the lipoid substance is at some distance from the external limiting membrane (*A*). The internal elastic membrane is insignificant (*B*) (Sudan III;  $\times 100$ .)

rabbit the expression of the lipoids was incomplete, because the contraction of the vessel was uniform (the elastic and muscle fibers are connected and dependent on one another—Benninghoff), and because the loosely arranged elastic fibers acted as a barrier to the expression as well as to the infiltration. In the peripheral vessels the irregularity of the contracting parts (the well developed elastic membrane contracts more forcibly than the remaining parts) and the absence of elastic barriers between it and the lumen effected a complete expression and may account for the absence of lipoids in the intima. It seemed strongly

suggested then that the deposit of lipoids in an artery was dependent not only on the infiltration of cholesterol but also on the expression of the same.

How can this mechanism be applied to the human being? The fibro-elastic layer of the intima of the aorta is markedly developed in the abdominal portion (fig. 3 *B*). In the thoracic portion this layer is characterized by a narrow dense fibro-elastic membrane (fig. 3 *A*). Correspondingly, the distance between the lumen of the aorta and the external limiting membrane of this layer increases from the thoracic to the abdominal portions. The internal elastic membrane is poorly developed throughout, while in the peripheral vessels it is markedly developed (Benninghoff, Nordmeyer).

With systole the lipoid-containing serum infiltrates into the entire arterial tree (Ribbert, Aschoff,<sup>e, f</sup> Hueck), and the lipoids may be arrested at the elastic barriers. The expression of the same will vary, depending on the factors mentioned.

In the thoracic portion the expression of lipoids will be more effective than in the abdominal portion of the aorta, because in the former the distance between the limiting membranes and the lumen is smaller. Also this membrane is denser and offers a more effective limitation to the infiltration as well as a more efficient expulsion. The fibro-elastic layer of the abdominal portion of the aorta corresponds somewhat to that of the rabbit's aorta in that a broad layer of loosely arranged elastic fibers is present between the lumen and the limiting membranes. Here the infiltration of lipoids extends among the loosely arranged elastic fibers and an effective barrier is reached only at some distance from the lumen. With diastole the loosely arranged fibers between the lumen and the external limiting membranes form a similar barrier to the out-flowing substance (fig. 7). This dissimilarity of structure may account for the greater frequency of atherosclerosis in the abdominal as compared with the thoracic portion of the aorta.

In the peripheral muscular vessels, where the internal elastic membrane is in proportion much better developed than in the aorta, with the diastolic recoil this membrane contracts forcibly and out of proportion to the other parts (the elastic fibers and the muscle fibers are two distinct systems in the muscular arteries—Benninghoff), thus effecting a more thorough expression. One has but to note the difference in the furrowing of the internal elastic membrane of muscular arteries in histologic preparations and to compare it with that of the aorta to be convinced of the inequality of contraction of the various layers of an artery.

In the arterioles, where the internal elastic membrane is practically absent, a deposit of lipoid and hyalin was noted in human beings, especially when hypertension was present (Gull and Sutton, Volhard, Rühl).

Now the question arises: Why is the lipoid deposit found to only a slight extent in persons up to 25 years of age, being even then reversible, while in older persons the fatty deposit increases rapidly and is non-reversible (Aschoff<sup>a</sup>)? This question is asked in the light of the fact that the histologic structure of the aorta under 25 years is not much different from that over this age.

In young as in old persons the mechanism of infiltration is probably not much different.

In nursing infants, in young persons at puberty, in women during pregnancy, and in persons who have passed through certain infectious diseases (especially typhoid fever—Ophüls), owing to a relative increase in the blood cholesterol or a disturbance of its mechanism, a deposit of cholesterol often occurs in the aorta, as the equilibrium between infiltration and expression is disturbed. With the recession of the cholesterol disturbance lipoids disappear from the aorta as a result of the compensated expression and the activity of the histiocytes (Langhans' cells) and because the fat is not "bound" to the interstitial substance or the elastic lamellae. With age two additional factors enter: (1) the aging of colloids which may bind or precipitate the fat (Bürger, Thannhauser, Wells, Aschoff<sup>a</sup>) and (2) the stretching or the disappearance of the elastic fibers preventing complete expression.

But not all people age at a similar rate and, as has been shown experimentally, the endowment of the elastic tissue may vary with different persons as well as in a given subject. Finally, with changes of the colloid and decreased elasticity of the vessels an arteriosclerosis does not necessarily follow, as will be shown later. True enough, a senile arteriosclerosis may develop with age with even a slight fat deposit due to degenerative changes, but this emphatically is not arteriosclerosis, and the latter is the more prevalent and more dreaded occurrence.

How is one going to determine the degree of aging in a vessel? From a histologic standpoint this is practically impossible in the early stages, as there is no method of determining the aging of colloid histo-chemically. Incubating aortas with sodium hydroxide (up to 20 per cent) or acetic acid (from 5 to 20 per cent) until a definite softening has taken place grossly does not show any alterations microscopically (Camac).

It is generally agreed that stretching of the elastic membranes and their replacement by fibrous tissue with age cause a dilatation of the vessel, and this process supposedly begins after the ascending stage of life (33 years, Jores, E. Kaufmann, Rössle, Aschoff,<sup>c</sup> Wells, Benninghoff, Bramwell, Duguid). As in the early stages of this process no microscopic changes are found, one has only the size of the vessel to rely on. In a large group of cases some insight into the degree of aging might be had by this method of comparison. It is admitted that this is a crude procedure, but one has none better at hand.

Table 23 presents the three measurements of the aorta for the average groups of males and females of both races. A progressive increase with age was noted for all cases (Jaffé and Sternberg, Rössle, L. Kaufmann, Suter). For the white race the highest percentage of dilatation was found at the level of the diaphragm (58 and 65 per cent in males and females, respectively). The greatest dilatation for the colored race was found at the bifurcation of the aorta (60 and 69 per cent, respectively). Some degree of error in the third measurement may be present, for when calcification occurs, the vessel may become pipestem-like and narrow. As this is not a frequent occurrence and was present in both white and colored races with equal frequency, the possibility of error is not great, considering the number of cases examined.

For the average group, embracing all factors that may involve the aorta (age, blood pressure, cholesterol metabolism, etc.), there was on

TABLE 23.—*The Measurements of the Aorta of the White and Colored Races from 25 Years Onward*

Age	Cases	White						Colored								
		Male			Female			Male			Female					
		Measurements, Mm.			Measurements, Mm.			Measurements, Mm.			Measurements, Mm.					
Age	Cases	1st	2nd	3rd	Cases	1st	2nd	3rd	Cases	1st	2nd	3rd	Cases	1st	2nd	3rd
25-30	5	57	39	32	10	53	34	29	8	62	40	30	19	57	39	29
31-40	24	65	46	37	20	59	39	30	26	66	45	33	16	63	43	33
41-50	52	72	49	39	29	66	47	35	21	68	46	40	15	67	50	38
51-60	52	75	53	42	21	66	47	37	15	75	54	45	10	76	52	37
61-70	40	78	57	44	17	75	54	40	8	76	55	47	4	79	60	49
71-	25	82	60	42	13	75	56	40	5	76	59	48	4	79	60	49
% increase		52	58	40		42	65	39		23	48	60		37	54	69

the average a 50 per cent increase in the size of the vessel throughout. Since physiologic growth is practically complete at the end of the ascending stage of life, this dilatation must be ascribed mainly to a degenerative process (Aschoff <sup>d, e, f</sup>).

If age were the only factor involved in the production of atherosclerosis, one would expect that aortas showing similar changes of age (as denoted by dilatation) should contain a similar amount of fat. That this is not the case is seen in table 24 A, B and C, where the measurements of the aorta are given according to the severity of the atherosclerosis, and in which cases are noted in which marked dilatation of the aorta occurred without atherosclerosis (table 24 A). True that the number of such cases decreases with age, and table 25 shows that there are no smooth aortas after 70 years, but it is significant that there are many present at 60 years (up to 27 per cent) and that they may be found even between 61 and 70 years. On the contrary, severe atherosclerosis occurred in persons in whom comparatively little dilatation of the vessels had occurred.

If it were possible to conceive that the other factors influencing atherosclerosis, such as a disturbance in the cholesterol metabolism and an increased blood pressure, could be staved off, the number of smooth aortas, irrespective of their aging, would probably be greatly increased.

It would be of great importance if in a similar way the degree of aging could be ascertained in various diseases, but here the other influencing factors are such that they may determine the size of the dilata-

TABLE 24.—A. *The Width of the Ascending Aorta in Various Conditions*

Age	White								Colored										
	Male				Female				Male				Female						
	S	SM	MS	AA	EH	sH	S	SM	MS	AA	EH	sH	S	SM	MS	AA	EH	sH	
A. The Width Above the Aortic Cusps																			
25-30	57			60	55	53		53	60		68	60	60	56	60		60	60	
31-40	64	68	68	68	67	71	58	65	68	74	65	64	68	69	69	70	65	66	
41-50	73	72	70	72	71	73	63	65	69	67	65	69	65	74	69	75	63	67	
51-60	71	75	77	76	79	77	66	67	65	66	63	67	68	77	76	77	86	72	
61-70	83	77	79	78	78	77		74	76	75	70	74	78	73	76	75	75	75	
71-		80	88	80	81	81		76	73	74	77	71		87	69	76	72	77	
B. The Width at the Diaphragm																			
25-30	39			42	38	34			34	41			37	37	41	38	42	42	42
31-40	45	47	51	49	49	50	39	40		43	50	40	42	44	53	47	49	45	41
41-50	49	49	52	49	49	49	44	46	51	48	50	48	49	49	45	48	47	47	46
51-60	49	52	53	54	56	54	46	48	51	48	48	47	52	50	63	52	57	49	48
61-70	53	57	56	57	60	56			52	56	54	52	53	55	58	63	57	60	55
71-		58	62	60	62	59			54	58	57	60	56	68	56	59	62	61	
C. The Width at the Bifurcation																			
25-30	32			36	31	29			29	31			28	28	31	28	30	30	30
31-40	36	36	40	39	40	38	31	32		34	39	32	33	37	40	38	40	35	36
41-50	38	36	42	40	39	38	35	37	39	37	37	40	40	38	40	39	40	39	42
51-60	39	43	43	43	44	36	37	43	38	37	37	40	41	54	42	37	38	33	38
61-70	43	44	44	44	49	41			38	42	40	44	41	49	41	54	48	48	40
71-		47	47	46	45	49			38	41	40	42	41		55	47	50	60	49
																	48	50	49

S = smooth.

SM = slight to moderate atherosclerosis.

MS = moderate to severe atherosclerosis.

AA = all cases of atherosclerosis.

EH = essential hypertension.

SH = without hypertension.

tion of the aorta and the measurements would be misleading unless carried out in a very large group. It is sufficient to say that in the supposedly degenerative diseases (diabetes mellitus, carcinoma, tuberculosis) there was no greater dilatation of the aorta, or aging, than was found in the average (also L. Kaufmann). Surprisingly, with essential hypertension, when sufficient cases were available (table 24 A, B and C), the width of the aortas was only slightly over that of the smooth aortas (Jaffé and Sternberg) and in many instances was equal to it (Kani). When the cases of atherosclerosis with hypertension and without hypertension were compared (table 24 A, B and C) the width was usually slightly increased in favor of the former (also Scheel, Jaffé and Sternberg) but not always so.

Figure 8 illustrates what has already been said. In this graph the widths of smooth aortas, those showing moderate to severe atherosclerosis and those in cases of hypertension in the white males were plotted, as this group comprised the greatest number of cases and represented the average trend of all groups.

Under the first measurement of the aorta, the width of the smooth ones excelled that of aortas presenting moderate to severe atherosclerosis

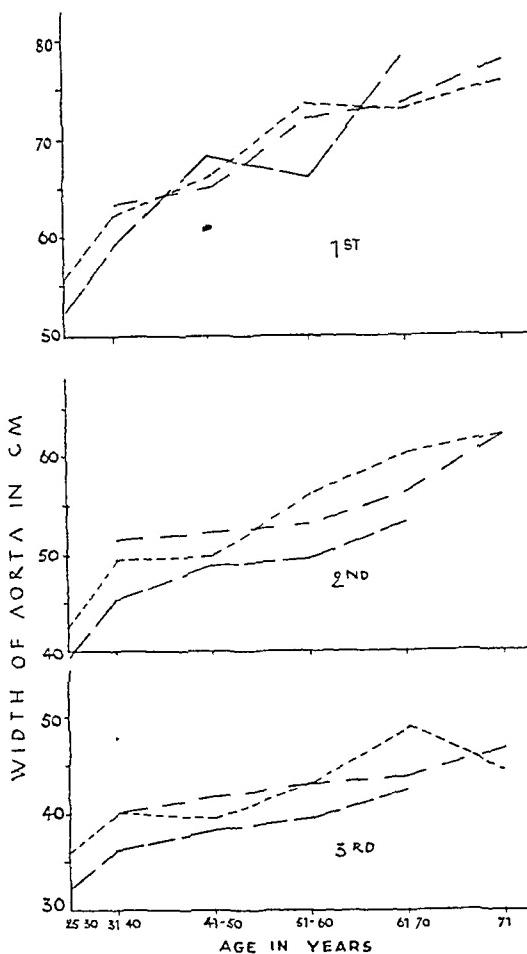


Fig. 8.—Measurements of the aorta in various conditions in different age groups of white males. Smooth aortas (— —); aortas showing moderate to severe atherosclerosis (— .. —) aortas from persons with essential hypertension (---).

and that of aortas of persons with hypertension, between 41 and 50 years and 61 and 70 years. Indeed, at from 61 to 70 years the width of the smooth aorta was greater than that of the atherosclerotic aorta at 71 years and over.

With the next two measurements, the width of the aorta ran somewhat parallel to the severity of atherosclerosis, but as atherosclerosis

is most prevalent in the abdominal portion of the aorta, other factors than age came into consideration. In the ascending portion of the aorta, where atherosclerosis is uncommon in all cases, a more uniform basis of comparison is available, and supposedly only the changes of age are there represented.

**Summary.**—From a histologic study of the arteries of the cholesterol-fed rabbit and the human being it is suggested that not only the infiltration of lipoids influences the latter's deposit in the arteries but that the expression of the lipoids is of equal importance.

The degree of development of the elastic limiting membranes influences both the infiltration and the expression. From a physical standpoint the latter is dependent on the nature of the elastic membranes,

TABLE 25.—*The Percentage of Cases With and Without Atherosclerosis According to Age, Sex and Race*

Age	White						Colored					
	Male			Female			Male			Female		
	No.	% cA	% sA	No.	% cA	% sA	No.	% cA	% sA	No.	% cA	% sA
25-30	5	0	0.0	100.0	10	0	0.0	100.0	9	2	22.2	77.8
31-40	24	9	87.5	62.5	20	3	15.0	85.0	26	11	42.2	57.8
41-50	52	26	50.0	50.0	29	19	65.6	34.4	21	7	41.6	58.4
51-60	52	38	73.0	27.0	19	12	63.1	36.9	15	11	78.9	21.1
61-70	40	37	92.5	7.5	17	17	100.0	0.0	8	6	75.0	25.0
71-	25	25	100.0	0.0	13	13	100.0	0.0	5	5	100.0	0.0

No. cA = number of cases with atherosclerosis.

% cA = percentage of cases with atherosclerosis.

% sA = percentage of cases without atherosclerosis.

the distance between the limiting membrane and the lumen of the vessel, and the inequality of the contraction of the various parts.

To demonstrate the foregoing statements, the following evidence was given: In the muscular type of arteries in the rabbit as in man, where the internal elastic membrane is well developed, there was from little to no lipoid deposit in the intima. In one muscular artery, where the internal elastic membrane had given way, a marked diffuse lipoid deposit occurred. When a moderate amount of lipoid was deposited among the elastic fibers of the rabbit's aorta it was situated, to a great extent, on the distal and not on the proximal aspect of the fiber. In the arterioles, where the internal elastic membrane is poorly developed or absent (spleen, kidney, liver), a lipoid deposit was again occasionally noted.

In the human being the development of the fibro-elastic layer of the abdominal portion of the aorta is marked, while in the ascending portion it is comparatively slight. This is considered to be the result

of a physiologic process in the ascending stage of life dependent on the greater circulatory demands made on the abdominal portion of the aorta.

A comparative study of the measurements of the aorta from birth to 30 years for the white and colored races revealed that there was no difference in the widths of the aortas for all cases up to 11 years. From then on the female aorta did not progress as rapidly as the male, especially the abdominal portion. The latter observation was explained by the difference in physical activity of the sexes after 11 years or thereabouts.

The abdominal portion of the aorta for both sexes and races increased in width to a higher degree (25 per cent) than the ascending portion. This difference is offered as an explanation of the variation in the development of the intima.

The fibro-elastic layer of the abdominal portion of the aorta, because of its marked development and its loosely arranged elastic fibers, serves as a barrier for the infiltration as well as for the expression of lipoid substances. This fact may probably account for the greater incidence and severity of atherosclerosis there.

The infrequency of lipoid deposition in the intima of the muscular arteries is explained by its strongly developed internal elastic membrane, which effects a more thorough expression of infiltrating substances. The poorly developed elastic membranes of the arterioles may account for their predisposition to hyaline and lipoid deposit.

In the ascending stage of life, when a disturbance of the lipoid metabolism takes place, as demonstrated by a relative increase of the blood cholesterol, the equilibrium between infiltration and expression is disturbed in favor of the former. When the hypercholesterolemia subsides, the expression component becomes compensated for and the cholesterol disappears from the aorta.

In adult persons, and more particularly in the descending stage of life, the aging of the colloids as well as the decreased elasticity of the vessel favors a binding or precipitation of the lipoids as well as a decreased expression of the same with the resultant irreversible cholesterol deposit.

Judging the aging of an aorta by the degree of dilatation, it was found that aging is a factor of first importance in the development of atherosclerosis. Aging in itself, however, does not lead to an atherosclerosis but to a pure senile sclerosis. The latter's occurrence is much less frequent than the former's.

Aortas in degenerative diseases, such as diabetes mellitus, tuberculosis and carcinoma, did not show any greater degree of dilatation than was found in smooth aortas. In essential hypertension the degree of dilatation was somewhat higher than that found in cases without hypertension.

B. THE BEARING OF HYPERTENSION ON THE INCLINATION TO  
ATHEROSCLEROSIS (F.A.A.)

That arterial tension influences the development of atherosclerosis is a long established fact (Jores, Aschoff,<sup>a, b</sup> Marchand, E. Kaufmann). Pointed examples are seen in the high incidence of changes in the pulmonary artery following increased pulmonary tension or in congenital stenosis of the aorta (Sternberg, Moschcowitz, Rosenthal). Whether the increased tension hastens the aging of the vessel or whether it favors the infiltration of cholesterol will be discussed later.

Experimentally, the development of atherosclerosis in cholesterol-fed animals is hastened by the addition to the diet of a substance elevating the blood pressure, such as epinephrine (Anitschkow,<sup>c</sup> Schmidtmann, Pfleidner). Increased blood pressure alone will not lead to a true athero-

TABLE 26.—*The Heart and Body Weights in All Cases Examined*

Age	Cases	White				Colored			
		Male		Female		Male		Female	
		Heart Wt., Gm.	Body Wt., Lbs.						
25-30	5	276	114	10	284	117	8	327	112
31-40	24	395	128	20	282	119	26	385	132
41-50	52	396	129	29	342	119	21	373	132
51-60	52	379	126	21	335	105	15	361	131
61-70	40	372	119	17	374	122	8	434	135
71-	25	416	140	13	340	112	5	419	132
								4	355
									146

sclerosis of the aorta in rabbits (Rühl,<sup>b</sup> Nuzum, Seegal, Garland and Osborne, Anitschkow<sup>c</sup>).

That atherosclerosis of the aorta is always accompanied by hypertension (Moschcowitz,<sup>a, b</sup>) is open to question (Hasenfeld, Hirsch, Lange<sup>b, c</sup>).

As a rule the weight of a heart and the blood pressure run parallel (Aschoff,<sup>e</sup> Lange,<sup>b, c</sup> Gewert). For the material presented it was found that a hypertrophy of the heart and thus hypertension could be considered when the heart weight was 400 Gm. or over for the male, and 375 Gm. or over for the female (excluding diseases of the heart proper). This weight was taken with all the blood removed and about from 1 to 3 cm. of the aorta attached. Naturally, lower values are obtained if the heart is prepared after the method of W. Müller, as was carried out by Rössle, Lange and also Levine and Carr, working in this laboratory. For a large material the method here employed carries but a small percentage of error (Gewert).

In figure 9 and table 26 the heart weights are given according to age, sex and race. For all cases the heart weight increased until the age from 40 to 50 and then became inconstant (also Fahr,<sup>b</sup> Müller, Rössle,

L. Kaufmann, Gewert). Although the body weight increased along with the heart weight for a general group, the relative heart weights also increased with age (Müller, Fahr, Rössle). As the present study was concerned only with the blood pressure as determined by the heart weight, the relative heart weights are not recorded (cf. Levine).

In comparing the heart weights with the fat angles of the aorta (F.A.A.) there was a certain amount of paralleling which suggested an influence of increased arterial tension (part I C).

In dividing the cases according to the severity of the atherosclerosis (table 27 A, B and C) it was noted that the weight of the heart increased

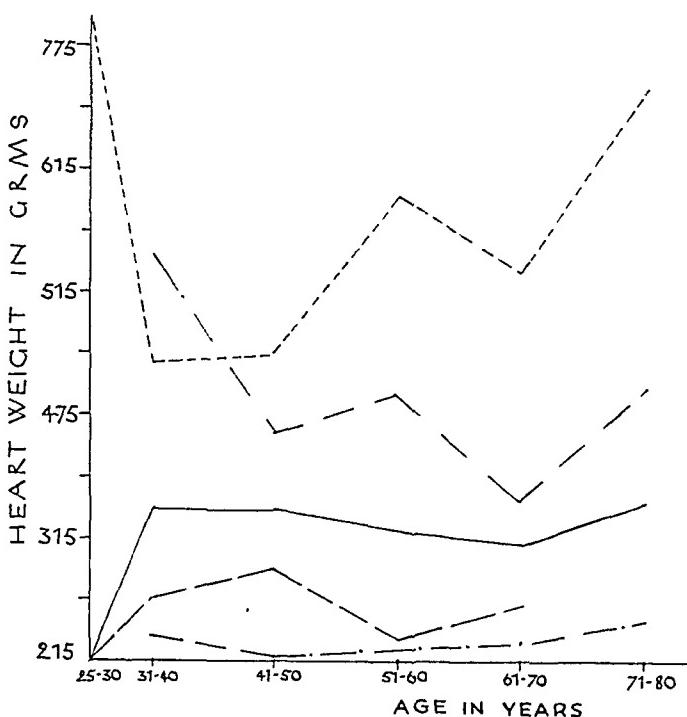


Fig. 9.—Heart weights corresponding to aortas in various conditions in different age groups of white males: average (—); smooth aortas (— —); aortas showing moderate to severe atherosclerosis (— . . —); aortas from persons with essential hypertension (- - -); aortas from persons with carcinoma (— . —).

with the severity of the atherosclerosis. However, when the groups were considered individually and the heart weight compared with the F.A.A. (part I B), such a parallelism was not so striking.

To have a larger number of cases for comparison, all cases of atherosclerosis were grouped together (table 28). Here one found that although the F.A.A. was about equal for all groups, the heart weights were decidedly different. Thus the heart weights from 25 to 50 years were usually greater than those past 50 years but the inclination to atherosclerosis was similar. Increased blood pressure, as determined

by the size of the heart, and the intensity of atherosclerosis are thus not directly proportional.

Under smooth aortas the heart increased in weight until 50 years (in the white female up to 60 years). From there on the heart became

TABLE 27.—*The Heart and Body Weights Corresponding to the Three Groups of Aortas*

Age	Cases	White				Colored			
		Male		Female		Male		Female	
		Heart Wt., Gm.	Body Wt., Lbs.	Cases	Heart Wt., Gm.	Body Wt., Lbs.	Cases	Heart Wt., Gm.	Body Wt., Lbs.
A. Smooth Aortas									
25-30	5	276	114	10	284	112	7	315	116
31-40	15	324	124	17	269	118	15	353	130
41-50	26	349	125	10	243	109	14	352	129
51-60	14	285	114	7	329	158	4	333	161
61-70	3	320	117				2	375	123
71-									
B. Aortas Showing Slight to Moderate Atherosclerosis									
25-30									
31-40	6	476	129	2	235	101	8	400	139
41-50	20	437	131	14	318	135	11	368	126
51-60	26	382	135	40	311	109	10	385	116
61-70	20	357	112	9	339	117	4	402	128
71-	11	310	117	5	370	136	2	402	119
C. Aortas Showing Moderate to Severe Atherosclerosis									
25-30									
31-40	3	600	146	1	640	112	3	503	109
41-50	6	461	149	5	552	136	2	465	147
51-60	12	494	136	4	433	103	1	230	158
61-70	17	404	126	8	418	118	2	460	111
71-	14	492	153	8	322	118	3	460	141

TABLE 28.—*The Relationship of the Heart Weight and the F.A.A. in All Cases of Atherosclerosis*

Age	Cases	White				Colored						
		Male		Female		Male		Female				
		Fat, Gm.	F.A.A., Degrees	Heart Wt., Gm.	Cases	Fat, Gm.	F.A.A., Degrees	Heart Wt., Gm.	Cases	Fat, Gm.	F.A.A., Degrees	Heart Wt., Gm.
25-30												
31-40	9	0.363	59.8	517	3	0.932	57.5	370	11	0.339	57.8	362
41-50	26	0.563	61.2	442	19	0.484	57.2	427	10	0.474	56.5	429
51-60	38	0.643	57.3	421	14	0.736	60.8	324	16	0.550	53.0	374
61-70	37	0.856	59.0	377	17	0.893	60.2	374	6	1.207	67.2	455
71-	25	1.036	60.5	424	13	1.189	62.9	353	5	1.289	64.8	437

smaller (Fahr, Rössle, L. Kaufmann; the Gewert normal values decreased in size after 40 years). The females of both races had smaller hearts than the corresponding males. The normal heart weight of the colored male was somewhat greater than that of the white male.

More positive evidence demonstrating the influence of hypertension on atherosclerosis is seen in table 29, in which only the cases of essential hypertension are included. The inclination to atherosclerosis lay between 60° and 65°, which was interpreted as a moderate to severe involvement of the aorta. For every age group, this inclination was greater than for the average group.

Similarly, chronic glomerular nephritis, because it was usually accompanied by an increased arterial tension, had also a higher incidence of atherosclerosis (cf. part II). As the degree of hypertension in nephritis was usually much lower than that of essential hypertension, the F.A.A. was correspondingly smaller. The small number of cases prevented the construction of a table, but the heart weights were as much as 30 per cent higher for this group than for the average.

Thus far the interconnection between atherosclerosis and hypertension has been stressed. In table 30, the cases of atherosclerosis without hypertension are listed. The heart weights were about similar to those with the smooth aortas and in some instances were smaller. An evaluation of the incidence of atherosclerosis without hypertension (table 30) proved that somewhat less than half the cases were not associated with an increased arterial tension.

By reversing the percentage of the cases without atherosclerosis in table 30, the incidence of hypertension with atherosclerosis could be deducted. In the white male hypertension played its greatest rôle up to 60 years, whereas in the white female the factor was most prominent after 60 years (effects of the climacteric). For the colored male there was a more or less equal distribution with age, while for the colored female hypertension was most marked between 41 and 50 years.

Drawing their conclusions from sources similar to those of the foregoing observations Jaffé<sup>b</sup> and Shapiro reported that the incidence of malignant nephrosclerosis was about five times as frequent in the colored race as in the white. The colored female was the most susceptible. The data given by Jaffé are as follows: 0.92 and 1.3 per cent for the male and female, respectively, of the white race, and 5 and 7.9 per cent for the colored male and female, respectively. The average age of colored persons with hypertension was 42 years. Hypertension in general was more prevalent in the colored than in the white race.

The foregoing evidence testifies to the importance of hypertension in the augmentation of atherosclerosis, but it shows equally that atherosclerosis can occur without hypertension (also Lange, Hasenfeld, Hirsch).

A slight diversion is here in order to examine the bearing of obesity on atherosclerosis and also on hypertension. There is no proof that overnutrition or undernutrition leads to atherosclerosis (Weiss and

TABLE 29.—*The Relationship of the Heart and Body Weights to the F.A.A. in Hypertension*

Age	White						Colored														
	Male			Female			Male			Female											
	F.A.A., Fat, Gm.	Avg. De- F.A.A., Degrees Gm.	Heart Wt., Lbs.	F.A.A., Fat, Gm.	Avg. De- F.A.A., Degrees Gm.	Body Wt., Lbs.	F.A.A., Fat, Gm.	Avg. De- F.A.A., Degrees Gm.	Heart Wt., Lbs.	F.A.A., Fat, Gm.	Avg. De- F.A.A., Degrees Gm.	Body Wt., Lbs.									
25-30.....	0.140	42.9	22.9	760	139 (1)*	... ..	0.659	72.6	25.9	640	112 (1)	0.460	65.0	44.2	76.4	44.3	560	140 (1)			
31-40.....	0.512	67.5	39.8	512	130 (2)	... ..	0.593	62.4	49.0	502	125 (7)	0.602	62.9	39.5	53.3	134 (3)	0.729	67.2	60.4	603	110 (8)
41-50.....	0.365	49.0	48.5	533	137 (8)	... ..	1.00	67.9	52.8	433	111 (3)	0.642	57.4	48.2	423	88 (4)	0.718	60.0	50.8	57.0	135 (1)
51-60.....	1.111	69.9	50.8	649	141 (6)	... ..	0.487	43.2	60.2	510	217 (1)	1.233	67.6	61.8	523	162 (4)	....	....	....	....	104 (2)
61-70.....	0.924	61.2	57.6	584	132 (5)	... ..	0.863	54.8	62.9	375	129 (5)	0.981	58.0	64.8	560	145 (1)	1.609	69.3	61.4	40.5	161 (2)
Average.....	60.2	46.7	..	..	60.2	50.1	..	..	..	..	..	..	..	..	..	..	..	..	..	..	..
							64.7	49.5	..	..	..	..	..	..	..	65.2	52.8	..	..	..	..

\* Figures in parentheses is the number of cases.

Minot), but it is generally conceded that obesity and hypertension exist together (Joslin, Dunham, Hunter and Rogers, O'Hara, Herrich, cited by Wyckoff).

In table 27 A, B and C, in which the body weights are given according to the severity of the atherosclerosis, there is no striking difference in the body weights of these groups. Thus the relative heart weights would necessarily be higher in the severer cases of atherosclerosis (also Gewert).

In the hypertension groups the body weights as a whole are greater than they are in the average groups (tables 29 and 26, respectively), but they cannot be considered as obese. It must be borne in mind that the cases examined were drawn from a poor class of people. When sufficient cases were available, the body weight on the average was

TABLE 30.—*The Relationship of the Heart and Body Weights in Cases of Atherosclerosis Without Hypertension*

Age	White										Colored									
	Male					Female					Male					Female				
	Cases		Without Hypertension		Gm.	Cases		Without Hypertension		Gm.	Cases		Without Hypertension		Gm.	Cases		Without Hypertension		Gm.
	Athero-sclerosis	No.	%	Heart Wt., Gm.	Body Wt., Lbs.	Athero-sclerosis	No.	%	Heart Wt., Gm.	Body Wt., Lbs.	Athero-sclerosis	No.	%	Heart Wt., Gm.	Body Wt., Lbs.	Athero-sclerosis	No.	%	Heart Wt., Gm.	Body Wt., Lbs.
25-30																				
31-40	9	1	11.1	350	121	3	2	66.6	235	101	11	5	46.4	295	127	7	3	42.8	278	126
41-50	26	11	42.3	297	112	19	8	42.1	251	126	7	4	57.1	277	140	11	2	18.1	315	96
51-60	38	17	44.6	297	120	12	9	75.0	291	104	11	6	54.6	321	126	8	5	62.5	293	121
61-70	37	22	59.4	313	111	17	8	47.0	250	111	6	2	33.3	295	93					
71-	25	14	56.0	329	125	13	4	30.7	284	113	5	2	40.0	355	114					
			48.1					48.4					47.5							36.1

below 140 pounds (63.5 Kg.). The lowest value was 88 (39.9 Kg.) and the highest 217 pounds (98.4 Kg.). The relative heart weights in the groups with essential hypertension are higher than in any other group (Levine and Carr, Gewert).

Compared with clinical observations, the blood pressure in an average group of white persons of the Western Hemisphere increases with age up to about 60 years (Romberg,<sup>a, b</sup> Volhard, Gager, Blackford, Bowers and Baker, Donnison, and others). However, what is considered as the average increase for the white civilized race is not the average for other races. The maximum normal blood pressure at 60 years was placed at 160 systolic and 100 diastolic by Romberg. For the colored people of eastern Africa it is 105 systolic and 67 diastolic (Donnison); for the Eskimos it is 129 systolic and 76 diastolic (Thomas). Low values were also reported for the Indians (Bengal) by Rogers, for the Chinese by Tung and Foster, for the Japanese by Rubner (quoted by Raab), and for the Mohammedans by Ruffer and Ismail.

Within the folds of the white race, the so-called normal blood pressure may in reality be abnormal. Saile has clearly shown that monks who live under different environments and diets have marked variations in their blood pressure. In one group who were strict vegetarians and spoke little the average blood pressure rarely went over 120 mm. of mercury (24 per cent), while in other monks who lived on an average diet, going among their parishioners, the blood pressure was practically always over 120 (70 per cent). The "normal" blood pressure of the white race in late adult life more likely borders on hypertension and may partly account for the high incidence of aortic changes.

It does not follow that a lower blood pressure excludes the possibility of atherosclerosis. As has been shown, in 48 per cent of these cases no inclination to an increased arterial tension was found. The Indians examined by Rogers and the Mohammedans examined by Ruffer have a lower blood pressure than the white race and supposedly a similar incidence of atherosclerosis. This would be compatible but it does not disprove the importance of hypertension.

Similar analogies can be found in studying the various diseases, considering as before that the "average" tables embrace equally the average possibilities that may lead to atherosclerosis.

In acute infectious diseases the cholesterol metabolism was considered as not appreciably altered. For the white race the F.A.A.s were everywhere similar except between 25 and 30 years. In comparing the respective heart weights (table 31) it is evident that for this group the weight was much lower than for the average group. Similarly, for the colored race differences were found which could be explained on the basis of the blood pressure (heart weights).

On the contrary, in carcinoma in which the heart weights were much lower than the average (also Gewert), the F.A.A.s were usually either equal to or slightly less than the average. Notwithstanding the fact that the heart weights were lower than those for the hearts with smooth aortas (F.A.A., 19.3°), the average F.A.A. for carcinoma was 43.1° (table 32). Increased blood pressure was certainly not an outstanding feature in this series.

In tuberculosis a relationship similar to that in carcinoma was found. Because of the lack of a sufficient number of cases a table was not made. The heart weights were as much as 25 per cent lower than for the average group. Other factors that increase blood pressure must be at work to effect the atherosclerosis.

With diabetes the F.A.A. was 59.7° or about 20 per cent greater than the average (48.8°). The heart weights for diabetes were as a rule about 20 per cent less than for the average. Here again the arterial tension played a minor rôle.

TABLE 31.—*The Relationship of Acute Infectious Diseases to the Heart Weight and the F.A.A.*

Age	White						Colored					
	Male			Female			Male			Female		
	Cases	Heart Wt., Gm.	Avg. Heart	Cases	Heart Wt., Gm.	Avg. Heart	Cases	Heart Wt., Gm.	Avg. Heart	Cases	Heart Wt., Gm.	Avg. Heart
25-30	2	212	276	2	252	294	12.0	19.6	1	340	327	24.3
31-40	6	382	395	10	267	282	30.6	34.0	4	331	385	46.2
41-50	9	407	396	4	262	342	46.3	48.4	7	333	373	46.0
51-60	5	417	379	3	280	335	46.2	51.2	8	351	452	48.2
61-70	6	331	372	3	488	337	46.8	58.4	2	375	359	41.4
71-	5	407	416	1	380	340	57.8	61.4			355	56.9

TABLE 32.—*The Relationship of Carcinoma to the Heart Weight and the F.A.A.*

Age	White						Colored					
	Male			Female			Male			Female		
	Cases	Heart Wt., Gm.	Avg. Heart	Cases	Heart Wt., Gm.	Avg. Heart	Cases	Heart Wt., Gm.	Avg. Heart	Cases	Heart Wt., Gm.	Avg. Heart
25-30	5	292.0	395	2	230	282	31.0	34.0	2	320	327	49.5
31-40	10	267.5	396	7	248	342	38.5	48.4	4	105	403	17.0
41-50	16	383.0	379	6	210	335	37.3	51.0	1	295	373	45.2
51-60	11	286.0	372	3	285	373	57.5	58.4	270	361	315	40.2
61-70	4	304.0	416	3	235	340	61.3	61.4	1	355	419	31.3
				Average....			43.1	50.1				34.5

M & F = male and female.

*Summary.*—Increased arterial tension plays an important rôle in the development of atherosclerosis, and when the former is present, the latter will invariably follow. Hypertension without atherosclerosis can occur—in the cases studied, to the extent of 7 per cent. Atherosclerosis, however, can develop without increased blood pressure—to the extent of about 48 per cent in the series examined.

In essential hypertension and chronic glomerular nephritis, in which the blood pressure is high, the F.A.A. is far above the average (65 and 55.9 per cent, respectively). In acute infectious diseases the F.A.A., when it equaled the average F.A.A., was associated with a similar heart weight, and if the F.A.A. was lower, the heart weight was lower.

In carcinoma and tuberculosis, in which the heart weights were much lower than the average and were more nearly equal to the weights of the hearts with the smooth aortas, the F.A.A. was not lower than the average and in some instances was higher.

In diabetes mellitus the F.A.A. was much higher than the average, although the heart weights were lower than the average.

It follows that factors other than increased blood pressure must be present to incite the development of atherosclerosis.

Body weight and atherosclerosis showed no interconnection. In hypertension the body weight was somewhat greater than the average, but obesity was rarely present.

#### IV. PATHOGENESIS OF ATHEROSCLEROSIS OF AORTA AS BASED ON CHOLESTEROL METABOLISM, BLOOD PRESSURE, AND INFILTRATION AND EXPRESSION OF LIPOIDS

From chemical, experimental and pathologic studies, and from opinions gathered from the literature, it appears that the pathogenesis of atherosclerosis of the aorta is principally dependent on age, cholesterol metabolism and blood pressure. Other factors, such as heredity, nervous influences and glands of internal secretion, owe their importance to the manner in which they influence the aforementioned ones. As this paper is limited to the pathogenesis of atherosclerosis, the further remote factors are not discussed in great detail.

##### NORMAL CONSIDERATIONS

There is a great difference of opinion as to when the atherosclerotic process begins. According to Anitschkow and his school (Stuckey, Chalatow,<sup>b</sup> Wolkoff<sup>b</sup>), the hyperplasia and hypertrophy of the internal elastic membrane are dependent on the lipoid deposit in the aorta from infancy onward.

In a comparative study of the width of the aorta from the first year of life to 30 years (ascending stage of life) in both sexes of the white and colored races it was found that the average widths were similar until 11 years of age. After that time, the female aorta developed less rapidly. The progression of growth corresponded closely to the difference in physical activity of the two sexes that begins at about this age. It could only be inferred that the demands placed on the circulation determined the size and also the structure of the vessel (also Benninghoff).

The normal histologic structure of the aorta at the end of the ascending stage of life varies in different parts of the vessel. In the ascending portion the intima is narrow and the fibro-elastic layer is compact. Extending to the abdominal portion of the aorta the intima becomes thicker, mainly through the proliferation of the fibro-elastic layer, the elastic fibers of which are loosely arranged.

The amount of fat deposited in the intima of the aorta before 25 years of age is exceedingly small (Görög) and is out of proportion to the proliferation described.

Under normal conditions, in the ascending stage of life the serum of the blood stream infiltrating through the inner two thirds of the aorta may show a hindering of its lipoids at the elastic barriers. With contraction of the vessel in diastole there is an expression of this substance. The mechanism of expression is dependent on the nature of the elastic barriers, the width of the intima and the inequality of the contracting parts (part III). The lipoid deposit is dependent on the disproportion of the infiltration over expression under normal conditions. It is for this reason that muscular arteries, which have markedly developed internal elastic membranes that contract more vigorously than the other constituent parts of the vessel, express the lipoids more efficiently. For a similar reason, the arterioles, having lost their internal elastic membrane, are more susceptible to lipoid and hyaline infiltration as seen best in hypertension.

When there is a disturbance of the cholesterol metabolism, as in infancy, puberty and pregnancy or after some acute infections, a cholesterol deposit in the aorta occurs, owing to a disturbance of the infiltration-expression equilibrium in favor of the former. After a subsidence of the hypercholesterolemia, the equilibrium is restored and the lipoid deposit disappears from the aorta (histiocytes also act as scavengers). Atherosclerosis of the aorta is thus rare in persons under 30 years of age.

#### AGE

After 30 years and more, particularly after 40 years, degenerative changes begin in the aorta as well as in other parts of the body. The

interstitial substance of the aorta ages as other colloids do with a decrease in hydration and a tendency to be transformed into a granular state (Wells). The elastic fibers, also being colloid masses, react in a similar way. These fibers decrease in number, and many are replaced by fibrous tissue, causing a dilatation of the vessel. Such changes having taken place, the serum cholesterol entering the aortic intima is deposited along the elastic barriers and remains there because the decreased elasticity prevents a thorough expression and favors stagnation, and because the dehydrated colloid favors a precipitation of the cholesterol esters as well as of calcium (Bürger and Schlemka, Wells).

The mechanism, however, requires other factors in addition to age, because without a disturbance of the cholesterol metabolism or an increased tension, an aorta may age without an appreciable lipoid deposit. This was clearly shown in a fair percentage of cases in which the dilatation of the aorta had taken place and yet the lipoid deposit was very small (part III). As the disturbances of the cholesterol metabolism as well as the increase in blood pressure are more prevalent with age, the incidence of atherosclerosis of the aorta also increases. It is significant that cases do exist in which physical colloidal changes have occurred and in which atherosclerosis is absent.

As one cannot measure the amount of aging of colloid in an aorta so is it impossible to determine the quality of the elastic tissue that one is endowed with. That heredity is an important factor is everywhere accepted (Jores, E. Kaufmann, Aschoff,<sup>e,f</sup> Janeway, T. DeWilliams). All other things being equal (environment, diet, etc.), the arteries of one person may age more rapidly than those of another. Similarly, in the same body, different vessels age at different rates (Wells, Wharton). For a large group of persons of the same race and sex living under like conditions the average aging of vessels was found to vary directly with age. That various diseases or malignant growths do not appreciably alter the degree of aging was shown in part III. One can then consider age as a variable constant in the development of atherosclerosis.

#### CHOLESTEROL METABOLISM

Whether disturbances of the cholesterol metabolism can be measured numerically only by the increase or the decrease of cholesterol in the blood, or whether there is an actual disturbance of the colloidal state of this substance, are questions yet to be answered. There is some evidence to show that variations in the colloidal properties of cholesterol esters can occur under similar temperatures (Chalatow<sup>a</sup>). Its determination in the body has not been definitely proved, so that at present the quantity of cholesterol in the blood is the only standard of measurement available. As the methods employed in the determination of the chole-

terol have been so numerous, the results are varied, and it cannot be definitely stated what the normal boundaries are.

Bürger found that normal persons show a marked hypercholesterolemia (100 per cent increase) after ingestion of 5 Gm. of cholesterol dissolved in 100 cc. of oil. The peak is reached in four hours, and after twenty-four hours the cholesterol returns to normal (contested by Barreda). Because other authors, using questionable methods of examining the blood and preparing the patient, have obtained negative digestive lipemias in man, the evidence that exogenous cholesterol is concerned in the pathogenesis of atherosclerosis has been discredited. Further, it is definitely known that although the endogenous cholesterol is not directly dependent on the exogenous cholesterol, the former is related to the latter.

If a relatively short digestive hypercholesterolemia occurs in persons who are accustomed to the ingestion of cholesterol-containing foods, most probably hypercholesterolemia of longer duration follows in persons who are not accustomed to this type of food. The latter possibility was suggested in the young colored person who on changing his mode of living and diet acquired a hypercholesterolemia, as was suggested by the early appearance of an arcus lipoides (part II). In such persons atherosclerosis sets in earlier than in the white people.

That the exogenous cholesterol may be of importance in atherosclerosis is also noted in comparative anatomy. The aves, particularly the parrot, are the only animals in which an atherosclerosis develops similar to that in man. The parrot is a meat and seed eater, substances containing cholesterol and neutral fats, respectively. It is admitted that age and the structure of the aorta of the parrot also play important rôles (Fox).

Endogenous cholesterol<sup>4</sup> may be influenced, as shown by hypercholesterolemia, in pregnancy, in retention of the cholesterol through blocking of the excretion of bile, in the transportation of cholesterol, as in inanition and the diseases leading to it (diabetes mellitus, tuberculosis, nephritis, carcinoma), in the liberation of cholesterol through cytolysis (acute infections, nephrosis, tuberculosis, carcinoma), by narcosis, and by glands of internal secretion.

How these factors act has been discussed elsewhere (parts II and V<sup>5</sup>). When a hypercholesterolemia does occur, the inclination to atherosclerosis, as determined by the fat angle of the aorta (F.A.A.), increases (part I). In cases in which only the cholesterol metabolism is affected, as in diabetes mellitus, the hyperlipcholesterolemia augments the inclination to

4. By endogenous cholesterol is meant that cholesterol that is not derived directly from the food.

5. Part V will be published in the December issue of the ARCHIVES.

atherosclerosis (F.A.A., 59.7°, as compared with the average, 45.5°). In the one case of lipoid nephrosis (a colored man aged 55) the F.A.A. was 62° (average, 48.2°). Although the incidence of atherosclerosis in diabetes is known to be high, in lipoid nephrosis the condition is not common. This is probably because nephrosis occurs as a rule in young persons and is not protracted over a long period.

What change occurs in the cholesterol metabolism with age is not known. Whether with age there is an actual increase in the blood cholesterol, or whether the digestive lipemia is accentuated, or whether a disturbance of the cholesterol metabolism occurs without an increase in the blood cholesterol is yet to be determined. Some authors have even reported a decrease in blood cholesterol in older persons. It must be remembered, however, that other conditions influencing the cholesterol metabolism also increase with age, such as diabetes mellitus, carcinoma and diseases in general. A slight disturbance of the cholesterol metabolism with age is more serious than a similar disturbance in youth in the development of atherosclerosis, as the "binding" of cholesterol is more effective with age.

How does cholesterol, especially its esters, affect the development of atherosclerosis? Aschoff,<sup>a, b, c</sup> Kawamura, Windaus and Schönheimer<sup>a, c, d</sup> have definitely proved that the cholesterol esters constitute the greatest part of the lipoid deposit in the aorta (57 per cent). This substance appears in the intima of the aorta long before degenerative changes can be seen with the microscope (Aschoff,<sup>a, b</sup> Zinzerling,<sup>a, b</sup> Görög). In histologic studies in human beings and cholesterol-fed rabbits it was found that the cholesterol esters stimulated histiocytic proliferation in the aorta as well as in the liver and kidney (Rosenthal; also McMeans, Stuckey, Wolkoff, Klotz and Manning). Anitschkow and his school described fibrous and elastic tissue proliferation as well as scar formation in the aortic intima after years of cholesterol feeding. Chalatow explained the cirrhosis of the liver that occurred in some cholesterol-fed rabbits on the basis of a change in the type of cholesterol, which then became more irritative (also Bailey<sup>c</sup>). What action the various cholesterol esters (palmitic, stearic and oleic) found in the blood and the aorta have is not known.

When one considers that the amount of fat in the aorta is almost directly proportional to the severity of the atherosclerosis (part I), it seems that the amount of fat determines the severity of the lesions and not vice versa. If one takes, e.g., fibrosis and scar formations in other parts of the body, the amounts of fat present in these lesions are usually small and depend as a rule on the necrosis of the constituents. In the aorta, although necrosis is also found and may accentuate the lipoid deposit, cholesterol esters are found long before the occurrence

of necrosis. It is true that a precipitation of the lipoids may occur as the result of aging of the colloid there (a submicroscopic phenomenon), but once the lipoid is deposited, it sets up an irritative action and probably accounts for the lesions that are known as atherosclerosis. When one considers that as much as 3 Gm. of fat may be deposited in the aortic intima, an irritation by the same is not difficult to conceive. Lipoids are found deposited to a slight degree in old dilated aortas, but here the senile sclerotic changes predominate. The latter condition is not atherosclerosis but senile sclerosis.

When cholesterol is deposited in an adult aorta at a rapid pace it effects the blocking of the blood supply to that part, and necrosis may follow. The latter in turn favors more deposit of lipoid as well as of hyalin and calcium (Wells, Barr). The intima covering a lipoid node undergoes pressure atrophy or necrosis. If the process is not rapid, the incrustation of calcium or hyalin in this atrophic intima follows and a calcific or hyaline plaque results. If the process of lipoid deposit is rapid, the overlying intima becomes necrotic and is swept away by the current of the blood stream, forming an atheromatous ulceration. As calcification is considered a secondary process, the mechanism of its deposition is not discussed at full length (for detailed studies, Barr, Wells and Schönheimer<sup>c</sup> may be referred to).

The older the subject the greater is the susceptibility to ulceration, while the younger the subject the greater is the inclination to fibrosis and hyalinization. In the latter, the expression mechanism is more efficient and the precipitation of the lipoid is less marked.

The sites of deposition of the cholesterol esters are to a great extent dependent on the structure of the intima and especially the elastic membranes. In the abdominal portion of the aorta the atherosclerotic process is more common, and this is accounted for by the loosely arranged elastic fibers of the fibro-elastic layer, which forms a barrier for the infiltration as well as for the expression of lipoids. Similarly, node and plaque formation is usually found about points where branches issue, because of a thickened intima there and a loose arrangement of the elastic fibers (Benninghoff). As in human beings, so in experimental animals, this has been found not only in the aorta but also where blood vessels divide, as at the division of the carotid artery.

The deposition of cholesterol is found in the same locations in youth as in age because of the peculiar structure of the aorta. Because in youth the infiltration and expression of lipoids is held in an equilibrium, and because the cholesterol is not "bound," atherosclerosis does not develop, while with age the "binding" of the cholesterol as well as the inefficient expression of the same accounts for the high incidence of atherosclerosis.

## BLOOD PRESSURE

The action of the blood pressure may be that of forcing the lipoids into the aorta at a greater pace (Aschoff,<sup>e</sup> Ribbert, Hueck, Stumpf) or that of hastening the aging process.

That increased pressure favors lipoid deposition is best seen from the infrequency of lipoid deposits in veins or in the pulmonary artery. With increase of the interpulmonary pressure, lipoid deposit in the pulmonary artery occurs readily (Sternberg, Moschcowitz,<sup>a, b</sup> Rosenthal) and similarly with increased pressure in the veins (Moschcowitz<sup>a</sup>).

In the greater circulation, increased blood pressure favors the development of atherosclerosis to a marked degree. The F.A.A. in essential hypertension was found to be 63° as compared with the average of 48°. There were some cases in which increased blood pressure did not lead to atherosclerosis. In a white man 30 years of age with an essential hypertension and a heart weight of 760 Gm., the amount of fat in the aorta was minimal (0.133 Gm.). However, the percentage of such cases was small (7 per cent). When an increase of the blood pressure had been prolonged, atherosclerotic changes of the aorta followed (Lange,<sup>b</sup> Moschcowitz<sup>a</sup>).

The question of determining the "normal" blood pressure is difficult. Thus, for persons of the white civilized race living under similar conditions, the blood pressure is much higher than for primitive races. Among the white civilized people, a change of environment and diet seems to be accompanied by a lowered blood pressure. Saile found that monks who are strictly vegetarian (excluding also eggs and butter) and who live entirely secluded (rarely talking) have a much lower blood pressure than monks who live on the average diet and go among their people. Similarly, Donnison found that the average blood pressure for the East African Negro (105 systolic and 67 diastolic at 60 years) was lower than that for the white race (140 systolic and 90 diastolic at 60 years). The colored race living in Chicago have a higher incidence of hypertension than the white race (Jaffé<sup>c</sup>).

Romberg<sup>b</sup> considers 160 systolic and 100 diastolic as the boundary of hypertension for the white race. This high "normal" value, in truth, should be considered as representing hypertension and may partially account for the high frequency of atherosclerosis in the white race.

The increased inclination to hypertension, especially of the malignant type, in the young colored persons dying at the Cook County Hospital may explain in part the increased inclination to atherosclerosis.

Local increases in blood pressure may also favor increased lipoid deposit. Thus, where vessels come in contact with bony parts, the resistance is greater and the deposit of lipoids is augmented. The

posterior wall of the aorta (Ophüls, Westenhöffer), the dural vessels (Lauda, Erdheim) and the internal carotid artery in the canal of the temporal bone are examples.

#### COMBINATION FORMS (BLOOD PRESSURE AND CHOLESTEROL METABOLISM DISTURBANCES)

It has been shown that marked disturbances of the cholesterol metabolism (diabetes mellitus and lipoid nephrosis) or marked increases of the blood pressure (essential hypertension) greatly increase the inclination to atherosclerosis. Combinations of these disturbances correspondingly alter this tendency.

In tuberculosis and carcinoma, in which there is a slight disturbance of the cholesterol metabolism but in which the blood pressure is lower than the average, the fat angle of the aorta (F.A.A.) is either lower than, or equal to, the average (part II). In acute infectious diseases, in which the cholesterol metabolism is only slightly disturbed for a short period of time, the F.A.A. is higher or lower, depending on the blood pressure. In chronic glomerular nephritis in which the cholesterol of the blood reaches a high "normal" or may be definitely increased, and in which the blood pressure is increased, the F.A.A. is increased. When both hypertension and marked cholesterol disturbances occur (diabetes mellitus with hypertension), the inclination to atherosclerosis is the highest (F.A.A., 76.8°; average, 48.5°).

The inclination to atherosclerosis is equally dependent on the cholesterol metabolism and hypertension, age being the variable constant. One cannot say that one or the other factor is capable of producing atherosclerosis alone, as the maintenance of blood pressure and cholesterol in the blood must always be contended with.

#### SUMMARY

The factors of age, cholesterol metabolism and blood pressure have been separately discussed in their relationship to atherosclerosis.

Although not all people age at a like rate, for a large homogeneous group living under similar conditions, the factor of age has been considered as a variable constant. Blood cholesterol and arterial tension also show a tendency to be augmented with senescence, but not necessarily. If it were possible to conceive that the aforementioned factors could exist alone, i.e., age, disturbance in the cholesterol metabolism and arterial tension, none could produce atherosclerosis alone.

Cholesterol, especially its esters, is of primary importance in atherosclerosis. Age and arterial tension check or augment the development of atherosclerosis. As it happens, age is inevitable and thus plays an exceedingly important rôle in the development of atherosclerosis, perhaps

as important as that of disturbances of the cholesterol metabolism and arterial tension combined. But atherosclerosis can occur in very young persons in whom a disturbance of the cholesterol metabolism exists without hypertension (as early as 10 years of age in persons with diabetes or lipoid nephrosis—Joslin). Hypertension alone will not lead to atherosclerosis, as has been shown experimentally and in the text. But as it is more prevalent in the descending stage of life it, too, plays a deciding rôle.

#### GENERAL SUMMARY

In a chemical analysis of 500 aortas of the white and colored races it was found that the fat content was not only directly proportional to age but also to the severity of atherosclerosis.

Because in a large group of cases the amount of fat in the aorta followed a regular ascent with age, the angle of inclination was designated as the fat angle of the aorta (F.A.A.), or the inclination to atherosclerosis. The deducted formula is:

$$\text{tangent F.A.A.} = \frac{(\text{fat in grams} \times 100)}{15} - 2.$$

The F.A.A. of all the cases studied was  $49.1^\circ$ , which was interpreted as a slight to moderate atherosclerosis.

Differences in the F.A.A. for the white and colored races and for the sexes suggested that the factor of age alone was not paramount in the production of atherosclerosis.

Because atherosclerosis occurred earlier in the colored race than in the white race, and because arcus lipoides was also prevalent in the former and not in the latter, it was deducted that the cholesterol metabolism may have played a rôle in the production of atherosclerosis. On the basis of the work of Joël, arcus lipoides occurring in young persons is associated with a hypercholesterolemia. To substantiate this point further it was found that atherosclerosis occurred more frequently in diseases in which a hypercholesterolemia was present (diabetes mellitus and lipoid nephrosis when long protracted). In carcinoma and tuberculosis, in which the cholesterol metabolism is less affected, the incidence of atherosclerosis was less. Furthermore, in compiling the literature relative to atherosclerosis among primitive races and in animals it was found that when atherosclerosis occurred, cholesterol and fats were present in the diet.

Measuring the aging of the aorta by its width showed that age played an important rôle in the development of atherosclerosis, but cases occurred in which aortic dilatation was not associated with atherosclerosis (up to 27 per cent).

It was inferred that age alone may lead to a senile sclerosis of the aorta but not to an atherosclerosis.

Arterial tension as determined by the weight of the heart was an important factor in the production of atherosclerosis, but atherosclerosis and hypertension are not synonymous. Indeed in 48 per cent of the cases, atherosclerosis occurred without hypertension.

In essential hypertension and in glomerular nephritis the F.A.A. was high, while in carcinoma and tuberculosis, in which the arterial tension was lower, the F.A.A. was lower but not correspondingly so. In acute infectious diseases the F.A.A. was proportional to the heart weights.

The development of the aorta from birth to 30 years was given, and it was inferred that the hyperplasia and hypertrophy of the intima were a functional response and not related to the fat deposit.

From morphologic and experimental evidence the deposit of lipoids was found to be closely related to the structure of the vessel and to the infiltration and expression of fats; the infiltration is dependent on the cholesterol in the serum, the blood pressure and the nature of the limiting membranes, while the expression of lipoids is dependent on the nature of the elastic barriers, the width of the intima and the inequality of the contracting parts.

The thickness of the intima of the abdominal portion of the aorta and the loose arrangement of the fibro-elastic layer were given as a partial cause for the higher incidence of atherosclerosis there.

The relatively thin intima and the well developed internal elastic membranes of muscular arteries were suggested as favoring expression of lipoids and therefore the rarity of lipid deposit in these vessels.

The reaction of the aortic intima to cholesterol esters in young adults is that of fibrous tissue proliferation and hyalinization, while in older persons ulceration is more prevalent.

#### LITERATURE

- Adler, I.: *J. Exper. Med.* **20**:93, 1914.  
Albutt, Clifford: *Diseases of the Arteries*, New York, The Macmillan Company, 1915.  
Alvarez, W. C.; Wiezen, R., and Mahoney, L. J.: *Arch. Int. Med.* **26**:381, 1923.  
Anitschkow, N.: (a) *Beitr. z. path. Anat. u. z. allg. Path.* **51**:379, 1913; (b) *Virchows Arch. f. path. Anat.* **249**:73, 1924; (c) *Experimental Arteriosclerosis in Animals*, in Cowdry, E. V.: *Arteriosclerosis*, New York, The Macmillan Company, 1933, p. 271.  
— and Chalatow, S.: *Centralbl. f. allg. Path. u. path. Anat.* **24**:1, 1913.  
Arndt, H. J.: *Ztschr. f. d. ges. exper. Med.* **54**:391, 1927.  
Aschoff, A.: *Ueber Entwicklungs-, Wachstums- und Altersvorgänge in den Gefässen*, Jena, Gustav Fischer, 1909.  
Aschoff, L.: (a) *Verhandl. d. deutsch. path. Gesellsch.* **10**:166, 1906; (b) *Beitr. z. path. Anat. u. z. allg. Path.* **47**:1, 1910; (c) *Lectures in Pathology*, New York, Paul B. Hoeber, Inc., 1924; (d) *Arch. ital. di sc. med.* **50**:1, 1927; (e) *Beihft. z. med. Klin.* **1**:1, 1930; (f) in Cowdry, E. V.: *Arteriosclerosis*, New York, The Macmillan Company, 1933.

- Bailey, C. H.: (a) Proc. Soc. Exper. Biol. & Med. **12**:68, 1914; (b) **13**:60, 1915; (c) J. Exper. Med. **23**:69, 1916.
- Barr, D. P.: Physiol. Rev. **12**:593, 1932.
- Barreda, P.: Klin. Wchnschr. **13**:290, 1934.
- Beitzke, H.: Virchows Arch. f. path. Anat. **267**:625, 1928.
- Beneke, R.: Beitr. z. path. Anat. u. z. allg. Path. **87**:285, 1931.
- Benninghoff, A.: Blutgefäße und Herz, Handbuch der mikroskopischen Anatomie, Berlin, Julius Springer, 1930, vol. 6, pt. I.
- Benson, R. D.; Smith, K. S., and Semenov, H.: Arch. Path. **12**:925, 1931.
- Beumer, H.: Ztschr. f. d. ges. exper. Med. **35**:328, 1925.
- Björling, E.: Virchows Arch. f. path. Anat. **205**:71, 1911.
- Blackford, J. M.; Bowers, J. M., and Baker, J. W.: J. A. M. A. **94**:328, 1930.
- Boinet, E., and Romary: Arch. de méd. expér. et d'anat. path. **9**:902, 1897.
- Bramwell, C.: Physical Properties of Arteries in Health and Diseases, in Cowdry, E. V.: Arteriosclerosis, New York, The Macmillan Company, 1933.
- Bürger, M.: Ergebni. d. inn. Med. u. Kinderh. **34**:583, 1928.
- and Habs, H.: Klin. Wchnschr. **6**:2221, 1927.
- and Schlamka, G.: Ztschr. f. d. ges. exper. Med. **55**:287, 1927.
- Camac, C. N. B.: Am. J. M. Sc. **129**:845, 1905.
- Cellina, M.: Cuore e circolaz **15**:506, 1931.
- Chalatow, S. S.: (a) Virchows Arch. f. path. Anat. **207**:452, 1912; (b) **272**:691, 1929.
- Chauffard, A.; Laroche, G., and Grigart, A.: Obstretique **4**:481, 1911.
- Richet, C., and Grigaut, A.: Compt. rend. Soc. de biol. **70**:317, 1911.
- Chuma, M.: Virchows Arch. f. path. Anat. **272**:691, 1929.
- Cirio, L.: Virchows Arch. f. path. Anat. **269**:739, 1928.
- Cramer, H.: Virchows Arch. f. path. Anat. **230**:46, 1921.
- Dewey, K.: Arch. Int. Med. **17**:575, 1916.
- DeWilliams, S.: Heredity Aspects of Arterial Hypertension in Relation to Arteriosclerosis, in Cowdry, E. V.: Arteriosclerosis, New York, The Macmillan Company, 1933, p. 537.
- Diehl, H. S., and Sutherland, K. H.: Arch. Int. Med. **36**:151, 1925.
- Donnison, C. P.: Lancet **1**:6, 1929.
- Duguid, J. B.: J. Path. & Bact. **29**:371, 1926.
- Erdheim, J.: Jahrb. f. Psychiat. u. Neurol. **39**:322, 1919.
- Fahr, T.: (a) Frankfurt. Ztschr. f. Path. **9**:15, 1911; (b) Virchows Arch. f. path. Anat. **239**:41, 1922.
- Foster, J. H.: Arch. Int. Med. **40**:38, 1927.
- Fox, W.: Arteriosclerosis in Mammals and Birds, in Cowdry, E. V.: Arteriosclerosis, New York, The Macmillan Company, 1933, p. 153.
- Freund, H., and König, W.: Arch. f. exper. Path. u. Pharmakol. **33**:317, 1928.
- Gager, L. T.: J. A. M. A. **99**:82, 1928.
- Gewert, M.: Veröffentl. a. d. Kriegs u. Konstitutionspath. **23**:1, 1929.
- Glasunow, M.: Virchows Arch. f. path. Anat. **261**:837, 1926.
- Görög, D.: Virchows Arch. f. path. Anat. **287**:603, 1933.
- Grigaut, M.: Le cycle de la cholesterinémie, Paris, Steinheil, 1913.
- Grundstein, N.: Arch. f. mikr. Anat. **47**:583, 1896.
- Gull, W., and Sutton, H.: Med.-Chir. Tr., London **55**:273, 1972.
- György, P.: Klin. Wchnschr. **3**:483, 1924.

- Hackel, W. M.: *Ztschr. f. d. ges. exper. Med.* **72**:762, 1930.  
Hesse, M.: *Virchows Arch. f. path. Anat.* **261**:225, 1926.  
Hirsch, C.: *Deutsche med. Wchnschr.* **38**:1817, 1913.  
Hoppe-Seyler, F.: *Medizinisch-klinische Untersuchungen aus dem Laboratorium für angewandte Chemie zu Tübingen*, Berlin, A. Hirschwald, 1866-1871, vol. 6, p. 593.  
Hueck, W.: *Beitr. z. path. Anat. u. z. allg. Path.* **66**:330, 1920.  
Hunt, H. M.: *New England J. Med.* **201**:659, 1929.  
Hurxthal, L. M.: *Arch. Int. Med.* **52**:86, 1933.  
Ignatowski, A.: *Virchows Arch. f. path. Anat.* **198**:248, 1909.  
Irsigler, F. H.: *Beitr. z. path. Anat. u. z. allg. Path.* **85**:221, 1930.  
Ismail, Abd-el. Aziz: *Lancet* **2**:275, 1928.  
Jaffé, R. H.: (a) *Klin. Wchnschr.* **10**:2081, 1931; (b) *Centralbl. f. allg. Path. u. path. Anat.* **55**:209, 1932.  
— and Sternberg, H.: *Med. Klin.* **51**:1311, 1919.  
Janeway, T.: *Clinical Study of Blood-Pressure*, New York, D. Appleton and Company, 1904.  
Joël, E.: *Klin. Wchnschr.* **3**:269, 1924.  
Jores, L.: *Arteriosklerose*, in Henke, F., and Lubarsch, O.: *Handbuch der speziellen pathologischen Anatomie und Histologie*, Berlin, Julius Springer, 1924, p. 703.  
Joslin, E. P.: *Ann. Clin. Med.* **5**:1061, 1927; *Ann. Int. Med.* **4**:54, 1930.  
Kani, I.: *Virchows Arch. f. path. Anat.* **201**:45, 1910.  
Kaufmann, E.: *Lehrbuch der speziellen pathologischen Anatomie*, Berlin, W. de Gruyter & Co., 1931, vol. 1.  
Kaufmann, L.: *Zur Frage der Aorta Angusta*, Inaug. Diss., Freiburg, 1919.  
Kawamura, R.: *Neue Beiträge zur Morphologie und Physiologie des Cholesterin-stoffwechsels*, Jena, Gustav Fischer, 1927.  
Key-Aberg, A.: *Ueber den Bau der Tunica intima der Aortenwand bei dem erwachsenen Menschen*, *Biol. Intersuch.* **1**:27, 1881.  
Klotz, O.: (a) *J. Exper. Med.* **8**:504, 1904; (b) *J. A. M. A.* **58**:1971, 1912; (c) *J. M. Research* **28**:157, 1916.  
— and Manning, M. F.: *J. Path. & Bact.* **16**:211, 1911.  
Knack, A., and Neumann, J.: *Deutsche med. Wchnschr.* **43**:901, 1917.  
Knauer, H.: *Klin. Wchnschr.* **8**:1745, 1929.  
Kolen, A. A.: *Virchows Arch. f. path. Anat.* **272**:679, 1929.  
Krause, K.: *Beitr. z. path. Anat. u. z. allg. Path.* **70**:121, 1922.  
Kuczynski, B.: *Klin. Wchnschr.* **4**:39, 1925.  
Labbé, M., and Heitz, J.: *Ann. de méd.* **18**:108, 1925.  
Lange, F.: (a) *Virchows Arch. f. path. Anat.* **248**:465, 1924; (b) *Hypertension in Relation to Arteriosclerosis*, in Cowdry, E. V.: *Arteriosclerosis*, New York, The Macmillan Company, 1933, p. 503.  
— and Webner, E.: *Deutsches Arch. f. klin. Med.* **160**:45, 1928.  
de Langen, G. D.: *Nederl. tijdschr. v. geneesk.* **65**:3056, 1921.  
Lauda, E.: *Beitr. z. path. Anat. u. z. allg. Path.* **68**:180, 1921.  
Lefkowitz, M., and Rosenberg, D.: *Frankfurt. Ztschr. f. Path.* **34**:174, 1926.  
Levine, V., and Carr, J.: *Arch. Int. Med.* **52**:429, 1933.  
Lotzmann, L. A.: *Virchows Arch. f. path. Anat.* **256**:117, 1925.  
Löwenthal, K.: *Verhandl. d. deutsch. path. Gesellsch.* **20**:137, 1925.  
Lubarsch, O.: *Ueber Arteriosklerose*, *Verhandl. d. Gesellsch. deutsch. Naturf. u. Aerzte*, Münster, 1912, vol. 84, p. 2.

- MacCallum, W. G.: Acute and Chronic Infections as Etiological Factors in Arteriosclerosis, in Cowdry, E. V.: Arteriosclerosis, New York, The Macmillan Company, 1933, p. 355.
- McCollum, E. V., and Simmonds, N.: The Newer Knowledge of Nutrition, New York, The Macmillan Company, 1929.
- McMeans, J. N.: J. M. Research **33**:475 and 481, 1916.
- and Klotz, O.: J. M. Research **34**:41, 1916.
- Marchand, F.: Verhandl. d. Kong. f. inn. Med. **21**:23, 1904.
- Mjassnikow, A. L.: Ztschr. f. klin. Med. **103**:267, 1926.
- Moissejeff, E.: Ztschr. f. d. ges. exper. Med. **60**:611, 1928.
- Moschcowitz, E.: (a) Am. J. M. Sc. **178**:244, 1929; (b) Virchows Arch. f. path. Anat. **283**:282, 1932.
- Müller, W.: Massenverhältnisse des menschlichen Herzens, Leipzig, Leopold Voss, 1883.
- Murata, M., and Kataoka, S.: Tr. Jap. Path. Soc. **7**:27, 1917.
- Neumann, R.: Frankfurt. Ztschr. f. Path. **42**:319, 1931.
- Newburgh, L. H., and Clarkson, S.: Arch. Int. Med. **31**:653, 1923.
- Newmann, E. T.: Arch. f. Schiffs- u. Tropen-Hyg. **34**:183, 1930.
- Nieberle, K.: Verhandl. d. deutsch. path. Gesellsch. **25**:291, 1930.
- Nordmeyer, N.: Beitr. z. path. Anat. u. z. allg. Path. **86**:149, 1931.
- Nuzum, F. R.; Seegal, B.; Garland, R., and Osborne, M.: Arch. Int. Med. **37**:733, 1926.
- Okunoff, N.: Virchows Arch. f. path. Anat. **259**:685, 1926.
- Ophüls, W.: The Pathogenesis of Arteriosclerosis, in Cowdry, E. V.: Arteriosclerosis, New York, The Macmillan Company, 1933, p. 249.
- Pallaske, G.: Frankfurt Ztschr. f. Path. **40**:64, 1930.
- Palmer, R. S.: J. A. M. A. **94**:694, 1930.
- Petroff, J. R.: Beitr. z. path. Anat. u. z. allg. Path. **71**:115, 1923.
- Pfleidner, E.: Virchows Arch. f. path. Anat. **284**:154, 1932.
- Pribram, H.: Prag. med. Wchnschr. **37**:205, 1912.
- Raab, W.: (a) Hormone und Stoffwechsel, Munich, Dr. F. P. Datterer & Cie, 1926. (b) Med. Klin. **14-15**:1, 1932.
- Ranke, O.: Beitr. z. path. Anat. u. z. allg. Path. **75**:269, 1926.
- Reuterwall, O. P.: Acta med. Scandinav. **55**:1, 1921.
- Ribbert, H.: Verhandl. d. deutsch. path. Gesellsch. **8**:168, 1904; Deutsche med. Wchnschr. **44**:953, 1918.
- Richter-Quittrner, M.: Wien. Arch. f. inn. Med. **1**:425, 1920.
- Rössle, R., and Roulet, F.: Mass und Zahl in der Pathologie (Pathologie und Klinik), Berlin, Julius Springer, 1932, vol. 5.
- Roger, L.: Glasgow M. J. **103**:1, 1925.
- Rohrschneider, W.: Virchows Arch. f. path. Anat. **256**:139, 1925.
- Romberg, E.: (a) Deutsche med. Wchnschr. **31**:1377, 1905; (b) Lehrbuch der Krankheiten des Herzens, Stuttgart, Ferdinand Enke, 1921.
- Rosenthal, F., and Holzer, P.: Arch. f. klin. Med. **135**:257, 1921.
- and Patrzek, F.: Deutsche med. Wchnschr. **45**:1037, 1919.
- Rosenthal, S. R.: Arch. Path. **10**:717, 1930.
- Rothschild, M. A.: Beitr. z. path. Anat. u. z. allg. Path. **60**:39, 1915.
- Rouzaud and Cabanis: Compt. rend. Soc. de biol. **74**:469, 1919.
- Rühl, A.: (a) Deutsches Arch. f. klin. Med. **156**:13, 1927; (b) Pharmakol. u. therap. Rundschau **140**:257, 1929.

- Ruffer, M. A.: Studies in Paleopathology of Egypt, Chicago, University of Chicago Press, 1921.
- Saile, F.: Med. Klin. **26**:929, 1930.
- Saltykow, S.: Beitr. z. path. Anat. u. z. allg. Path. **43**:147, 1908.
- Scheel, O.: Virchows Arch. f. path. Anat. **191**:135, 1908.
- Schmidtmann, M., and Huttich, M.: Virchows Arch. f. path. Anat. **267**:601, 1928.
- Schönheimer, R.: (a) Virchows Arch. f. path. Anat. **249**:1, 1924; (b) **251**:732, 1924; (c) Ztschr. f. physiol. Chem. **160**:61, 1926; (d) **177**:143, 1928; (e) Klin. Wchnschr. **43**:1793, 1932.
- Schultz, A.: Virchows Arch. f. path. Anat. **239**:415, 1922.
- Schultze, W. H.: Ergebn. d. allg. Path. u. path. Anat. **13**:253, 1909.
- Shapiro, P. F.: Arch. Int. Med. **48**:199, 1931.
- Shapiro, S.: J. Exper. Med. **45**:595, 1927.
- Soli, V.: Ann. di clin. med. **14**:251, 1924.
- Ssokoloff, N. A.: Virchows Arch. f. path. Anat. **245**:203, 1923.
- Ssolowjew, A.: (a) Virchows Arch. f. path. Anat. **241**:1, 1923; (b) **250**:359, 1924; (c) Ztschr. f. d. ges. exper. Med. **69**:94, 1930.
- Starkodamski, L. M.: Zur Frage ueber die experimentelle Arteriosklerose, Inaug. Dissert. St. Petersburg, 1909.
- Steinbiss, W.: Virchows Arch. f. path. Anat. **212**:151, 1913.
- Stepp, W.: Beitr. z. path. Anat. u. z. allg. Path. **69**:233, 1921.
- Sternberg, V.: Beitr. z. path. Anat. u. z. allg. Path. **82**:307, 1929.
- Stocks, Percy: Race and Climate as Possible Factors in Arteriosclerosis, in Cowdry, E. V.: Arteriosclerosis, New York, The Macmillan Company, 1933, p. 195.
- Strauch, C.: Beitr. z. path. Anat. u. z. allg. Path. **61**:536, 1916.
- Stucky, N. W.: Veränderungen der Kaninchenaorta unter dem Einfluss der Fütterung mit animalischer Nahrung, Inaug. Dissert., St. Petersburg, 1910; Centralbl. f. allg. Path. u. path. Anat. **21**:668, 1910.
- Stumpf, A.: Beitr. z. path. Anat. u. z. allg. Path. **59**:390, 1914.
- Suter, F.: Arch. f. exper. Path. u. Pharmakol. **39**:289, 1897.
- Sydenstricker, E.: Statistical Study of Arteriosclerosis, in Cowdry, E. V.: Arteriosclerosis, New York, The Macmillan Company, 1933, p. 131.
- Thannhauser, S. J.: (a) Deutsches Arch. f. klin. Med. **141**:290, 1922-1923; (b) Lehrbuch des Stoffwechsels und der Stoffwechselkrankheiten, Munich, J. F. Bergmann, 1929.
- Thérèse, L.: Rev. de méd. Paris **13**:123, 1893.
- Thölldte, M.: Beitr. z. path. Anat. u. z. allg. Path. **77**:61, 1927.
- Thoma, A.: Virchows Arch. f. path. Anat. **204**:1, 1911.
- Thomas, W. A.: J. A. M. A. **88**:1559, 1927.
- Torhorst, H.: Beitr. z. path. Anat. u. z. allg. Path. **36**:210, 1904.
- Tsunoda, T., and Umehara, N.: Tr. Jap. Path. Soc. **6**:90, 1916.
- Tung, Chen-Lang: (a) Chinese J. Physiol. **1**:93, 1928; (b) **4**:117, 1930.
- Versé, M.: (a) Beitr. z. path. Anat. u. z. allg. Path. **63**:789, 1916; (b) Deutsche med. Wchnschr. **51**:49, 1925.
- Virchows, R.: Wien. med. Wchnschr. **6**:809, 1856.
- Voigt, H.: Der Aufbau der normalen Aorta, Inaug. Dissert., Marburg, 1904.
- Volhard, F., and Suter, F.: Nieren und ableitende Harnwege, in von Bergmann, G., and Staehelin, R.: Handbuch der innerer Medizin, Berlin, Julius Springer, 1931, vol. 6, pt. 1.

- Wacker, L.: *Ztschr. f. physiol. Chem.* **80**:383, 1912.  
—and Hueck, W.: *Arch. f. exper. Path. u. Pharmakol.* **71**:373, 1912-1913.  
Weiss, S., and Minot, G. R.: Nutrition in Relation to Arteriosclerosis, in Cowdry, E. V.: *Arteriosclerosis*, New York, The Macmillan Company, 1933, p. 233.  
Wells, H. G.: The Chemistry of Arteriosclerosis, in Cowdry, E. V.: *Arteriosclerosis*, New York, The Macmillan Company, 1933.  
Westenhofer, P.: *Deutsche med. Wchnschr.* **48**:518, 1922.  
Wharton, A. S.: *Old Age, the Major Involution*, New York, Paul B. Hoeber, Inc., 1929.  
Widal, F.; Weil, A., and Laudat, M.: *Semaine méd.* **32**:529, 1912.  
Wilheim, R., and Fuchs, G.: *Biochem. Ztschr.* **247**:297, 1932.  
Windaus, A.: *Ztschr. f. physiol. Chem.* **67**:174, 1910.  
Wolkoff, K.: (a) *Virchows Arch. f. path. Anat.* **256**:751, 1925; (b) *Beitr. z. path. Anat. u. z. allg. Path.* **85**:386, 1930.  
Wyckoff, J.: The Treatment of Arteriosclerosis, in Cowdry, E. V.: *Arteriosclerosis*, New York, The Macmillan Company, 1933.  
Yuasa, D.: *Beitr. z. path. Anat. u. z. allg. Path.* **80**:570, 1928.  
Zeek, P.: *Am. J. M. Sc.* **184**:350, 1932.  
Zinzerling, W. D.: (a) *Virchows Arch. f. path. Anat.* **255**:676, 1925; (b) *Beitr. z. path. Anat. u. z. allg. Path.* **88**:241, 1932.

# Case Reports, Laboratory Methods and Technical Notes

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## HAND-SCHÜLLER-CHRISTIAN'S DISEASE AND TUBERCULOSIS

HUGH G. GRADY, M.D., AND HAROLD L. STEWART, M.D., PHILADELPHIA

Excellent reviews of the clinical, biochemical and morphological manifestations of Hand-Schüller-Christian's disease or, as Chester<sup>1</sup> designates the condition, lipoid-granulomatosis, have recently been published by Lichty,<sup>2</sup> Merritt and Paige,<sup>3</sup> Davison,<sup>4</sup> Pick,<sup>5</sup> Chiari,<sup>6</sup> Sosman,<sup>7</sup> Henschen,<sup>8</sup> Kleinmann<sup>9</sup> and Ighenti.<sup>10</sup> No attempt will be made to review the literature except as it pertains to the case under consideration. The features of particular interest in this case include changes in the lymph nodes, spleen and certain areas of the bone marrow resembling those in the group of nonlipoid histiocytosis described by Foot and Olcott.<sup>11</sup> In addition the lungs and mediastinal lymph nodes contained tuberculous foci which were frequently clean-cut and separate but which at other times exhibited some overlapping with the granulomatous lesions of xanthomatosis. The latter lesions were widespread, often incipient, just as specific as the tuberculous changes, and occurred in such unusual situations as the hypophysis, liver, spleen and lymph nodes. Death occurred under ether anesthesia, which fact, coupled

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From the Pathological Laboratories of the Jefferson Medical College and Hospital, the Jefferson Hospital Tumor Clinic and the Philadelphia General Hospital.

1. Chester, W.: *Virchows Arch. f. path. Anat.* **279**:561, 1930. Chester, W., and Kugel, V. H.: *Arch. Path.* **14**:595, 1932.
2. Lichty, D. E.: *Arch. Int. Med.* **53**:379, 1934.
3. Merritt, K. K., and Paige, B. H.: *Am. J. Dis. Child.* **46**:1368, 1933.
4. Davison, C.: *Arch. Neurol. & Psychiat.* **30**:75, 1933.
5. Pick, L.: *Am. J. M. Sc.* **185**:601, 1933.
6. Chiari, H.: *Verhandl. d. deutsch. path. Gesellsch.* **25**:347, 1930; *Ergebn. d. allg. Path. u. path. Anat.* **24**:396, 1931; *Virchows Arch. f. path. Anat.* **288**:527, 1933.
7. Sosman, M. C.: *Am. J. Roentgenol.* **23**:581, 1930; *J. A. M. A.* **98**:110, 1932.
8. Henschen, F.: *Acta paediat. (supp. 6)* **12**:1, 1931.
9. Kleinmann, H.: *Virchows Arch. f. path. Anat.* **282**:613, 1931.
10. Ighenti, W. K.: *Virchows Arch. f. path. Anat.* **282**:585, 1931.
11. Foot, N. C., and Olcott, C. T.: *Am. J. Path.* **10**:81, 1934.

with the changes in the suprarenal and thymus glands, emphasizes one of the risks involved when operative procedures are contemplated in persons with Hand-Schüller-Christian's disease.

#### REPORT OF A CASE

In a white girl aged 3 years, pain, tenderness and discharge from both ears developed in May 1931. A partial facial paralysis appeared on the left side and subsequently cleared up; later a complete and permanent paralysis developed on the right side of the face. She was treated with roentgen therapy, and a right-sided simple mastoidectomy was done. On microscopic examination of tissue removed from the mastoid process a diagnosis of sarcoma was made. A second biopsy specimen was examined by Dr. Arnold Rich of Baltimore who favored the diagnosis of Hand-Schüller-Christian's disease. A large nodule which appeared below the right ear in March 1932 and similar lesions developing in the left ear in June of the same year responded well to radium therapy. A left-sided simple mastoidectomy was done subsequently and revealed a condition similar to that encountered on the right side.

She was admitted to Jefferson Medical College Hospital on Oct. 18, 1932, with a mass of granulation tissue in the right posterior auricular region, bilateral open sinuses in the mastoid areas, a discharge from both ears and a right-sided facial paralysis. On exploring the left mastoid area, all landmarks, including the posterior osseous wall of the bony canal, were found to be destroyed, and the dura was covered with fibrous granulation tissue. A radical operation was done, and the tissue removed consisted of vascular cellular granulation tissue with many inflammatory cells, chiefly polymorphonuclear leukocytes and large mononuclear cells. No exophthalmos or other abnormality was noted on ophthalmologic examination on Jan. 17, 1933. On roentgenographic examination (Jan. 24, 1933) the middle fossa of the skull appeared depressed, and a sharply defined area of osseous destruction, about the size of a silver dollar, was demonstrated in the lower posterior portion of the right parietal bone. This bony defect, which had not increased in size by April 9, 1933, was incised and drained, and smears of the material obtained showed many pus cells, gram-positive cocci and gram-negative rods. *Staphylococcus albus* and *Streptococcus nonhaemolyticus* grew in cultures planted with this material. There was a slight degree of secondary anemia. The urine contained a faint trace of albumin on several occasions, but no records of the urinary output were kept. The blood contained 59 Mg. of sugar per hundred cubic centimeters (Benedict method; normal, from 60 to 100 mg. per hundred cubic centimeters); the blood cholesterol was not studied.

The enlarged cervical nodes regressed somewhat under roentgen therapy, but the other lesions and the patient's general condition remained about the same. Death occurred suddenly under ether anesthesia during an attempted tonsillectomy on June 8, 1933.

*Postmortem Examination (Six Hours After Death).*—The head was enlarged, and the face presented a square appearance. There were numerous superficial petechial hemorrhages in the skin of the abdomen and recent incisions lined by dark necrotic material along the angle of each jaw and above and behind the right ear. Purulent fluid was observed exuding from each external auditory meatus.

No gross lesions were observed in the brain. The dura in the immediate vicinity of the sella turcica contained several small yellowish nodules, and its inner surface was covered by a yellowish-brown, granular deposit which could be scraped away with ease. In the petrous portions of both temporal bones large areas were replaced by soft, necrotic, bright yellowish nodules.

The thymus was normal in size and position but practically completely replaced by irregular, firm, yellow, discrete and confluent nodules.

The left lung weighed 100 Gm. and measured 13 by 9 by 4 cm. In the upper outer portion of the lower lobe a circumscribed nodule of caseous yellow material 1 cm. in diameter was found, encapsulated by dense connective tissue which sent linear projections into the surrounding pulmonary parenchyma especially in the direction of the regional lymph nodes. A similar but smaller and less sharply circumscribed lesion was present in the apex of the upper lobe. The remainder of this lung and the entire right lung, which weighed 130 Gm. and measured 14 by 9 by 4 cm., were red and mottled by grayish flecks.

Several bronchial lymph nodes were converted into dry caseous masses surrounded by dense fibrous tissue. Other thoracic and many abdominal nodes were enlarged, apparently hyperplastic, flecked with reddish spots and softened in areas.

The spleen, weighing 210 Gm. and measuring 13 by 7 by 4 cm., was moderately firm and on section presented a bright red pulp studded with sharply circumscribed, grayish dots resembling follicles.

The liver weighed 680 Gm. and measured 21 by 13 by 5 cm. The gallbladder and bile ducts appeared normal. In general the hepatic tissue was pale, friable and translucent. In the outer aspect of the left lobe there was a cystic area 2 cm. in diameter, filled with thin, opaque bile-stained fluid and lined on the inner aspect by a brown ragged membrane. The outer wall of this lesion consisted of a thick layer of dense fibrous tissue which sent prolongations into the surrounding parenchyma and contained several small, firm, yellow nodules.

The suprarenal glands were thin. The heart, pancreas, gastro-intestinal tract and genito-urinary tract showed no noteworthy changes.

**Microscopic Examination:** **Lymph Nodes:** The follicles were decreased in number and the lymph cords were atrophic with alterations in cellular content but without much change in structural pattern. There was marked diminution in the number of follicular and medullary lymphocytes with corresponding replacement by plasma cells, eosinophilic polymorphonuclear leukocytes and another type of cell which will be referred to hereafter as a "pleochromatic histiocyte." The last-mentioned type of cell was round, polyhedral, elongated or spindle-shaped, averaging 20 microns in diameter with frequent variations between 15 and 40 microns. The nucleus was pale, vesicular, centrally placed and sometimes crumpled, with a sharply etched outline dotted with small granules of chromatin. A dark, violet nucleolus was often observed. The cytoplasm was dense, dull, homogeneous, rarely slightly vacuolated and usually neutrophilic but with frequent variations toward acid or basic reactions. A few small black granules were frequently present in osmic acid preparations, and occasionally one or two reddish granules and needle-shaped crystals were observed after staining with nile blue sulphate. Many of the crystals were doubly refractile when viewed with crossed Nicol prisms. The pleochromatic histiocytes were present within and around lymph sinuses and blood vessels, in large numbers within the follicles, diffusely in areas of granulation tissue, and in rosette formations throughout the medulla. In association with other cells they formed an important constitu-

ent of nodular cellular collections which were most numerous in the medullary portions of the node and which were present also immediately beneath the capsule, where many of them occupied the positions formerly held by the follicles. As a rule these nodular lesions consisted exclusively of closely packed or discretely separated pleochromatic histiocytes, loosely supported by a delicate reticulum (fig. 1). Others contained eosinophils, lymphocytes, plasma cells, erythrocytes, pigment and strands of fibrin, all of which were subject to the phagocytic action of the pleochromatic histiocytes. Young vascular, edematous and hemorrhagic granulation tissue was interspersed around these nodules and elsewhere in the lymph node, distorting the architectural pattern somewhat and being

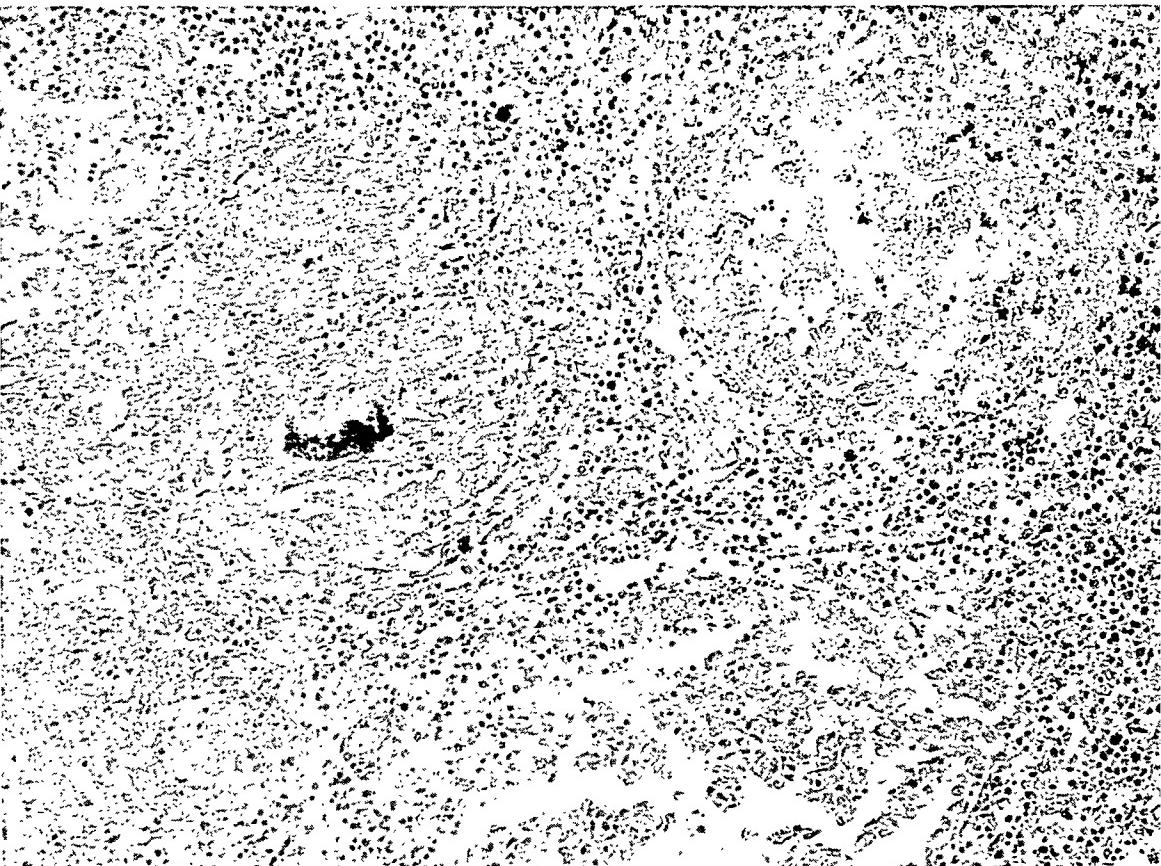


Fig. 1.—Bronchial lymph node showing two nodular cellular collections of xanthomatosis to the right and a small tubercle to the left. The nodule in the right upper part of the field is hemorrhagic and composed of loosely arranged pleochromatic histiocytes with engulfed pigment; reduced from  $\times 200$ .

especially compact and hyalinized about the smaller blood vessels. Minute areas of hemorrhage were found in the lymph sinuses containing pleochromatic histiocytes and outside the capsule of the node. The picture in some of the bronchial lymph nodes was further complicated by the presence of tuberculous lesions in varying stages of activity (fig. 1). One such area measuring 3 by 6 mm. was composed of a central caseous and calcific mass encapsulated by dense hyalinized connective tissue directly continuous with an area of granulation tissue which contained focal collections of cells. Several discrete and small confluent miliary tubercles which sometimes involved the nodular cellular collections were

present in another node. Remnants of eosinophils and pleochromatic histiocytes were recognized in the central caseous area of some tubercles, which regularly showed a high lipin content.

**Spleen:** The appearance under crossed Nicol prisms, the reactions to fat stains and the other splenic changes were essentially similar to those in the non-tuberculous lymph nodes. However, there was a more marked tendency to the occurrence of peri-arterial fibrosis and hyalinization, and the replacement of lymphocytes by pleochromatic histiocytes appeared to proceed from the center of the follicle toward the periphery. The pleochromatic histiocytes were especially numerous in the red pulp about the borders of malpighian corpuscles where they occurred in rosette formation and in characteristic nodular cellular collections. Numerous small hemorrhages were scattered throughout the pulp, and accumulations of histiocytes, eosinophils, monocytes and lymphocytes were present beneath the intima of many of the larger veins.

**Liver:** The hepatic cells were separated by moderately dilated sinusoids and showed fine and coarse cytoplasmic vacuolation especially in the outer portions of the lobule. The Kupffer cells were large and numerous with abundant, vacuolated cytoplasm containing small granules of brown to olive green pigment. The portal radicles were prominent owing to an increased fibrosis and a marked infiltration of lymphocytes without much increase in the bile ducts. Under nile blue sulphate stain large and small globules of neutral fat were demonstrated in most of the hepatic cells in the extreme outer border of the lobule where many small dark blue granules were present in both the hepatic and the Kupffer cells. Many needle-shaped crystals arranged singly or in sheaves and staining various shades between red and blue were scattered throughout the lobule in the sinusoids, in the perivascular tissue spaces and in Kupffer cells.

The cystic area, noted grossly, was separated from the surrounding hepatic parenchyma by a thick, fibrous wall lined on its inner aspect by a wide zone of young granulation tissue covered by flattened, compressed cells on the surface and sparsely infiltrated by polymorphonuclear leukocytes, eosinophils, lymphocytes, monocytes, plasma cells and foam cells. The cytoplasm of the foam cell was usually abundant and appeared as a fine network of reticulated character containing varying quantities of amorphous granules or compact masses of pale or refractile yellow pigment. As a rule a single, small round nucleus dotted with finely granular chromatin material was present, but two or three nuclei were not unusual, and as many as fifteen were occasionally observed in the larger cytoplasmic masses. Red droplets of neutral fat and reddish granules, greenish needle-shaped crystals and coarse irregular red or blue spicules were demonstrated in the cytoplasm of these cells stained with nile blue sulphate. Under crossed Nicol prisms great numbers of doubly refractile needle-shaped crystals were observed. This layer of granulation tissue gradually became devoid of cells and was succeeded by a wide band of dense hyalinized connective tissue containing several larger arteries, veins, bile ducts and nerve trunks, but showing no tendency to infiltrate the hepatic parenchyma except for a few coarse prolongations.

**Thymus:** The thymus consisted principally of granulation tissue interspersed with linear and branching compressed streaks of lymphocytes, reticular cells and a few corpuscles of Hassall, composed of concentric rings of hyalinized or more or less calcific material, practically devoid of nuclei. The peripherally placed cells in these streaks frequently contained coarse droplets of neutral fat, reddish granules and greenish needle-shaped crystals in nile blue sulphate preparations.

The granulation tissue was very vascular and practically devoid of lipins in the immediate vicinity of the compressed thymic tissue where it was apparently youngest. Elsewhere the capillaries tended to disappear and the small blood channels remaining were rendered conspicuous by their thickened walls and perivascular hyalinization. Foam cells similar to those in the xanthomatous cyst of the liver were at times the only constituents of the granulation tissue and were also found in the adventitia of the vessels, in the perivascular fibrous tissue and outside the capsule of the organ (fig. 2). At other times lymphocytes, plasma cells, monocytes, eosinophils and many vacuolated histiocytes were scattered diffusely in the

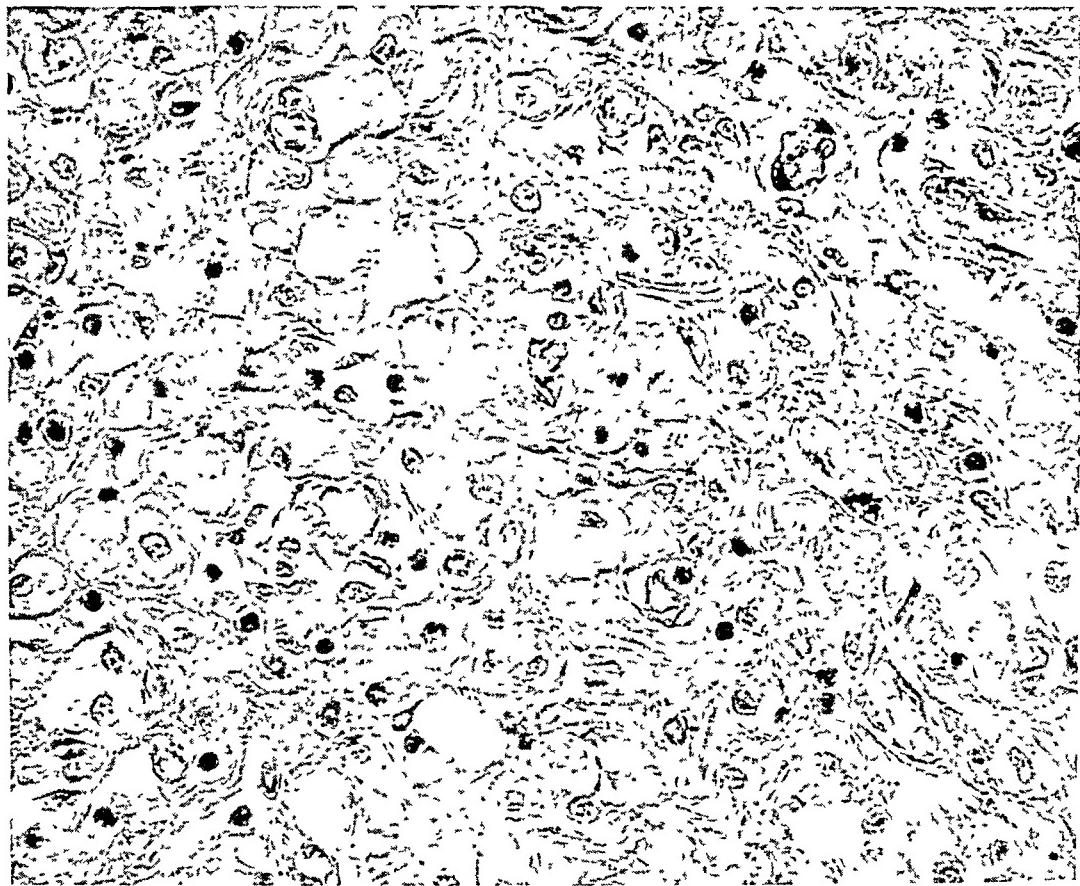


Fig 2.—Section of the thymus showing a xanthomatous area of closely packed foam cells supported by connective tissue stroma;  $\times 250$ .

meshes of the loose fibrillar reticulum. These cells also formed nodular cellular collections presenting a characteristic arrangement. The center was composed of monocytes, myelocytes and eosinophilic leukocytes densely packed together and surrounded in succession by (1) a narrow, clear border devoid of cells, (2) a cellular area containing extensively vacuolated pleochromatic histiocytes and several giant phagocytes supported by a loose fibrillar reticulum and (3) beyond this a syncytium of cells showing mitosis and mobilization. Transitional stages were observed between the foam cells and the pleochromatic histiocytes which became increasingly vacuolated. Round, refractile, acidophilic hyaline droplets were seen resembling those frequently observed in the renal tubular epithelium. Under crossed Nicol prisms great numbers of doubly refractile needle-shaped crystals were observed.

Lungs: The lumens of the alveoli and bronchi and the walls of the alveoli, bronchi and blood vessels contained varying numbers of lymphocytes, plasma cells, eosinophils, monocytes and especially pleochromatic histiocytes. Only an occasional foam cell was observed in the alveoli or lying against a septal wall. There were frequently a marked peribronchial and perivascular proliferation of granulation or hyalinized connective tissue which replaced large areas of pulmonary parenchyma, usually subpleural. These areas were studded with nodular cellular collections infiltrated by vacuolated histiocytes and surrounded by emphysematous pulmonary parenchyma. Old calcified tuberculous areas and discrete and small confluent miliary tubercles, similar to those in the lymph nodes, were also noted.

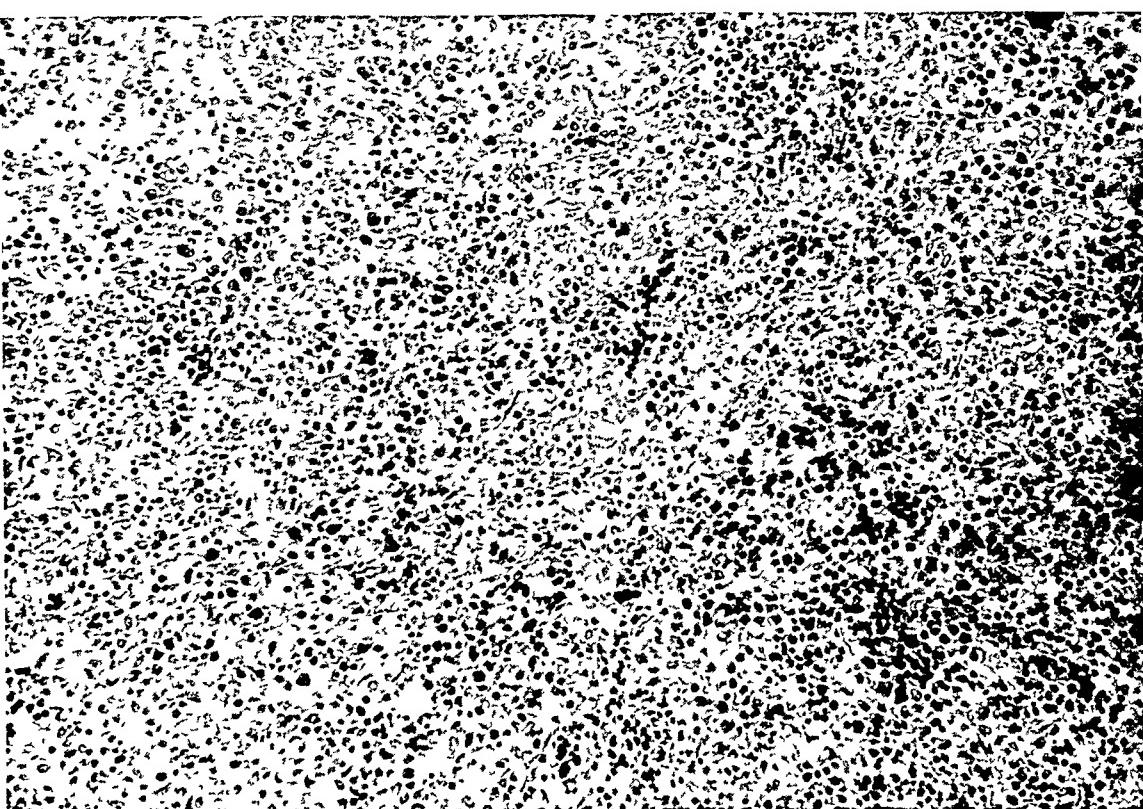


Fig. 3.—Bone marrow showing a gradual transition from hyperplastic marrow on the right to an area infiltrated by pleochromatic histiocytes which are difficult to distinguish from megakaryocytes; reduced from  $\times 200$

Hypophysis: The nerve cells in the posterior lobe were reduced in number and separated, especially near the pars intermedia, by a slight diffuse and focal infiltration of lymphocytes, plasma cells, eosinophils, foam cells and single or multinucleated pleochromatic histiocytes supported by granulation tissue. The remainder of the hypophysis showed no noteworthy changes.

Dura: The outer surface (periosteal layer) was lined by a continuous layer of granulation tissue containing pleochromatic histiocytes, foam cells, lymphocytes, eosinophils and plasma cells scattered diffusely or arranged in nodular cellular collections. There was a tendency for the occurrence of perivascular hyalinization and fibrosis about several nerves and pacchionian bodies. Under crossed Nicol prisms there were great numbers of doubly refractile needle-shaped crystals.

Bone: The periosteal margin of the petrous portion of the temporal bone was partially decalcified and very irregular owing to small indentations occupied by osteoclasts. Along this line there were marked erosion and numerous spicules, small flecks and thin irregularly shaped segments of bone lying partially detached and sequestrated. The interior of the bone showed advanced destruction and partial replacement by granulation tissue similar to and continuous with that on the dura.

The outer periosteal fibers of the sternum split at certain points to encapsulate small round or flat nodules of granulation tissue containing numerous accumulations of foam cells. The bone underlying these nodules was atrophied. Similar nodules were present immediately inside the marrow cavity of some of the vertebral bodies.

Bone Marrow: The bone marrow of the vertebrae, sternum, ribs and femur showed marked hyperplasia of all the elements, especially of the eosinophilic myelocytes. The megakaryocytes were numerous and showed many degenerating forms. Scattered generally throughout the myeloid tissue, but especially in and about the nests of primitive erythrocytes, were many large single or multinucleated cells with an abundance of smooth, homogeneous, deeply staining, slightly acidophilic cytoplasm, resembling somewhat that of the megakaryocytes (fig. 3). When numerous these cells tended to displace the normal bone marrow elements. Other cells were present which resembled these in many respects except that the cytoplasm showed varying degrees of vacuolation. A section through the marrow of the femur showed a gradual transition between an area of hyperplastic myeloid tissue and an area of young granulation tissue, containing a few foci of marrow cells and many plasma cells, monocytes and large pleochromatic histiocytes. There was marked fibrosis about the larger blood vessels. Cells with hyaline droplets like those observed in the thymus were present in the bone marrow.

Kidney: There were marked vacuolation and swelling of the tubular epithelial cells and a slight cellular reaction with pleochromatic histiocytes in the connective tissue of the pelvis.

Suprarenal Glands: There was atrophy of the medulla and to a less extent of the cortex which showed marked lipoid depletion.

Brain: Significant changes were not present.

#### COMMENT

In this case the lesions did not present the lipoid reactions or the morphologic characteristics of Niemann-Pick's disease, and they can readily be distinguished from those of Gaucher's disease by the absence of the typical Gaucher cell with its clear, wrinkled and longitudinally striated cytoplasm. The essential features were manifestly those of Hand-Schüller-Christian's disease and included granulomatous lesions, hemorrhages, necroses, ultimate fibrous cicatricial transformation, dural and periosteal granulations, xanthomatous cysts of the liver, hypoplasia of the suprarenal glands and characteristic lesions in the lungs, kidney, bone marrow, thymus and pars nervosa of the pituitary gland. The foam cell was present in typical form in many areas and resembled that of the secondary xanthoma (Plewes<sup>12</sup>). The principal lesions, viewed under crossed Nicol prisms, contained many doubly refractile needle-

12. Plewes, L. W.: Arch. Path. 17:177, 1934.

shaped crystals, and chemical analysis of tissue obtained from the dura, thymus and cyst in the liver showed a high content of cholesterol. In Cowie and Magee's<sup>13</sup> case the xanthomatous masses had a high content of total lipoid, 50 per cent of which was cholesterol. Hand-Schüller-Christian's disease is regarded as a disturbance of lipoid metabolism or of lipoid excretion resulting in the storage of lipoid substances in the granulomatous lesions of the various organs and tissues. There is phagocytic activity on the part of the cells of the reticulo-endothelial system which remove an excess of lipoids in certain areas in which infection or trauma, particularly of the bone and skin, may have stimulated a collection of histiocytes. Although infection has been shown to have some connection with the development of the disease, Sosman<sup>7</sup> emphasized the fact that it is frequently possible to obtain a history of preceding infection in the majority of diseases of childhood. Whether the disturbance in the lipoids and in the reticulo-endothelial system is due to an avidity of the cells for lipoids or is a compensatory mechanism resulting from a failure of proper fat metabolism is problematic.

The evolutionary type of process in Hand-Schüller-Christian's disease resembles that of lymphogranulomatosis, which occupies a borderline position between neoplasm and infection. These two conditions have been confused clinically and especially as the result of biopsy examination, but their manifestations are usually quite dissimilar, and the histologic pictures bear only a superficial resemblance to each other in atypical cases. Certain of the changes in the petrous portions of the temporal bones are suggestively similar to those of osteitis fibrosa cystica, a point stressed particularly by Snapper and Parisel.<sup>14</sup> Foot and Olcott<sup>11</sup> described a condition under the designation of nonlipoid histiocytosis in which the changes resembled those in the lymph nodes, spleen and parts of the bone marrow in our case. They collected from the literature a number of similar examples having in common this marked proliferation of lipoid-free histiocytes in association with eosinophils and plasma cells. Some of the cases were believed to be infectious in origin, others leukemic with aleukemic intervals, while those occurring in childhood regularly presented a marked generalized purpuric eruption of the skin and scattered small petechiae over the greater part of the body, leading to the diagnosis of thrombocytopenic purpura. The clinical picture was indefinite and poorly characterized, although a history of different types of recent infection was generally elicited and the changes in the lymph nodes were somewhat similar to those in nodes draining infected areas. It appears probable that a relationship may

13. Cowie, D. M., and Magee, M. C.: Arch. Int. Med. **53**:391, 1934.

14. Snapper, I., and Parisel, C.: Quart. J. Med. **2**:407, 1933.

exist between this group of so-called nonlipoid histiocytosis and at least some cases of Hand-Schüller-Christian's disease.

The marked overgrowth of pleochromatic histiocytes and the absence of foam cells in the lymph nodes, spleen and parts of the bone marrow are features of Hand-Schüller-Christian's disease which have not received general recognition. The cytoplasm of the pleochromatic histiocyte has been described as uniformly basophilic or eosinophilic with marked variations between these reactions (Robertson and Warren<sup>15</sup>). In our case it was usually basophilic in the lymph nodes, eosinophilic in the spleen and dura and pale and neutrophilic in the hypophysis; its other characteristics were quite uniform in the various organs and tissues in which it was studied. Its origin from the reticular cell of the splenic follicle was suggested by the fact that it appeared first in the center of the malpighian corpuscle. Some of its forms were difficult to distinguish from the megakaryocyte of the bone marrow and from bizarre types of multinucleated plasma cells in the spleen and lymph nodes. In the two latter situations only a small amount of lipoid material was stored in the pleochromatic histiocytes, although their phagocytic character was evidenced by the ingestion of pigment, débris and other cells. Transitional forms between these histiocytes and the foam cells in the dura, thymus, lungs and bone marrow were frequently encountered. The nodular cellular collections, composed of pleochromatic histiocytes alone or in association with other cells, represented a prevailing feature of all the granulomatous lesions of xanthomatosis. The foci composed exclusively of pleochromatic histiocytes had not been infiltrated by other cellular elements or the latter had undergone necrosis or been phagocytosed by the histiocytes. As the diffuse lesions of granulation tissue became less vascular and more compact and hyalinized with age, the walls of the remaining capillaries were thickened, and there were a perivascular deposition of dense collagen fibers and an infiltration of large single and multinucleated foam cells in certain situations, as in the thymus and dura.

The tuberculous foci in the lungs and lymph nodes were easily distinguished from the granulomatous lesions of xanthomatosis although there was a definite overlapping of the two in a few of the lymph nodes. In some instances miliary tubercles were engrafted secondarily on the nodular cellular areas, and remnants of pleochromatic histiocytes, eosinophilic leukocytes and plasma cells could be recognized in the caseous necrotic material. Frequently the pleochromatic histiocytes exhibited no apparent defensive reaction to the extension of the tuberculous process, but at other times they became radially arranged like epithelioid cells at the periphery of the tubercle; the cytoplasm tended

15. Robertson, S. H., and Warren, S.: Arch. Path. 15:193, 1933.

to remain pleochromatic, and the nuclei became enlarged and more rectangular without undergoing mitosis. The areas of xanthomatous granulation tissue in the lung appeared to be especially susceptible to involvement by tuberculous lesions. In the case reported by Chiari<sup>6</sup> the patient died of cavernous pulmonary tuberculosis without xanthomatous involvement of the thoracic or abdominal viscera.

The morphologic changes in the thymus and suprarenal glands coupled with the emergency type of death occurring under ether anesthesia, which had, however, been administered successfully three times previously, emphasizes the operative risks involved when surgical procedures are contemplated in patients with Hand-Schüller-Christian's disease. In our case the heart and blood vessels were not particularly hypoplastic, but the suprarenal glands were small and thin and the structural alterations in the thymus were advanced. A definite statement cannot be made regarding a lowered alkaline reserve, but a single determination showed that the blood sugar was at a low normal level. There is not sufficient evidence at present to state that these features may constitute a new syndrome resembling that of status lymphaticus which has no very definite pathologic basis itself (Marine,<sup>16</sup> Young and Turnbull<sup>17</sup> and Waldbott<sup>18</sup>). The various syndromes, such as diabetes insipidus, infantilism, exophthalmos and others occurring in the group of lipoid diseases, apparently depend on the prevailing type of lesion involving different organs and structures of the body, being incidental and not essential features of the condition.

#### SUMMARY

A case is presented of lipoid granulomatosis (Hand-Schüller-Christian's disease) with widespread involvement of the organs and tissues and with coexistent tuberculous lesions in the lungs and lymph nodes. Death occurred suddenly under ether anesthesia.

16. Marine, D.: Arch. Path. **5**:661, 1928.

17. Young, M., and Turnbull, H. M.: J. Path. & Bact. **34**:213, 1931.

18. Waldbott, G. L.: Am. J. Dis. Child. **47**:41, 1934.

## CARTILAGINOUS METAPLASIA IN AORTIC ATHEROSCLEROSIS IN A PARROT

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Fox<sup>1</sup> reports finding degenerative arterial disease in thirteen, or 1.8 per cent, of a series of birds of the order Psittaci. In his material medial degeneration and fibrosis of smaller vessels were more prominent, and the aorta was less often the principal site of the disease. Degenerative changes were more prominent than productive. Intimal proliferative plaques also occurred. Fox recorded no instance of cardiac hypertrophy or of myocardial scarring in Psittaci.

The bird which is the subject of this report was a large green female about 30 years old; it had been a household pet in one family for most of that time. It died with symptoms of diarrhea, and the carcass was sent to the National Institute of Health for examination as to the possible presence of psittacosis.

At autopsy the ascending and transverse aorta and the innominate artery were unusually thick and rigid, a change which extended to the subclavian arteries within the chest. The heart did not appear much enlarged, but was quite thick and muscular and was firmly contracted. Intestinal distention was the only other noteworthy gross observation.

Histologically the heart showed focal interstitial scars in the thickened musculature of the septum and left ventricle. The ascending and transverse aorta and the innominate artery presented a marked thickening of the intima irregularly encroaching on the musculo-elastic media. There were also a few focal interstitial fibrous scars in the markedly thinned media. The intima was basically fibrous. It contained a few focal areas of coarse vacuolation in which there were cellular necrosis and slight granular calcium deposition. There were also scattered clumps of needle-like clear spaces embedded in fibrous tissue, probably representing cholesterol crystals. The most striking change was the extensive replacement of the intima by irregular masses of typical hyaline cartilage. With the Giemsa stain the usual metachromasia of cartilage was present, but nonmetachromatic areas were often interspersed between the metachromatic pericorporeal zones.

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From the National Institute of Health.

1. Fox, Herbert: Disease in Captive Wild Mammals and Birds, Chicago, J. B. Lippincott Company, 1923.

Stains for fat showed large numbers of singly refractile globules staining intensely with Sudan IV lying apparently in the cartilage cell spaces, and in the matrix, masses of doubly refractile material, part of which was tinged with Sudan IV, lying in fibrous areas between the fibers. Nile blue sulphate showed no fatty acids, but many of the globules in the cartilage were doubly refractile and divided into quadrants by a dark cross.

The descending thoracic aorta presented only an eccentric rarefied and vacuolated fibrous intimal plaque. The abdominal aorta, the pulmonary artery and its branches and the coronary, renal, splenic and hepatic arteries were normal. In accordance with the usual practice in presumably infected birds, the carcass was incinerated immediately after evisceration; hence, no material from the alar or femoral vessels was available.

#### SUMMARY

A case of atherosclerosis of the thoracic aorta in a parrot is reported. The sclerotic intima presented a minor amount of degenerative changes and an extensive cartilaginous metaplasia.

## General Review

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### PATHOGENESIS OF POSTPRIMARY PROGRESSIVE TUBERCULOSIS OF THE LUNGS

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CHICAGO

For many years the interest of the pathologists was devoted to the changes of fully developed pulmonary tuberculosis, and many classifications were suggested which were based on the anatomic findings. Microscopic investigations disclosed the most minute details of the histogenesis, progression and regression of the specific reactions to the tubercle bacilli, and little additional information in these fields could be expected. At present the discussion of the pathologists centers about the point of origin of the progressive pulmonary tuberculosis of the postprimary period and about the rôle which exogenous superinfection and endogenous hematogenous infection of the lungs play in causing the isolated, progressive tuberculosis of this organ.

The studies of Birsch-Hirschfeld, von Baumgarten, Schmorl, Abrikosoff and Aschoff seemed to have so firmly established the apical beginning of pulmonary tuberculosis (*phthisis pulmonum*, Aschoff) that other possibilities received very little attention from the pathologists. The clinicians, however, were less unanimous in the assumption that progressive pulmonary tuberculosis always started in the apex. Fishberg stated that long before the use of x-rays two British authors, W. Ewart and J. Kingston Fowler, referred to the frequent subapical location of early tuberculous consolidations. With the introduction of roentgen rays in the diagnosis of pulmonary lesions the significance of the apical changes in tuberculosis became doubtful. According to Sweany, Cook and Kegerreis, Gekler was the first to champion the idea that roentgenologic findings pointed toward an infraclavicular location of the earliest lesions in pulmonary tuberculosis. Following the publications of Wessler and especially the fundamental studies of Assmann, numerous clinical and roentgenologic reports have established that progressive pulmonary tuberculosis most frequently starts acutely with a lesion below the clavicle.

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The new conception of the great significance of the early infra-clavicular infiltrations was bound to cause much concern among the orthodox believers in an apical beginning of pulmonary phthisis. To the morbid anatomists the findings of the roentgenologists were of limited value since the apical lesions might be so small as to require microscopic search. At a convention of specialists in tuberculosis in 1928, Graeff made the statement that there was no definite proof against the classic conception of the morbid anatomists that pulmonary tuberculosis, as a rule, started in the apex.

Fully developed pulmonary tuberculosis yields no information as to its probable point of origin. It may be recalled that judging the age of a tuberculous lesion from its morphologic appearance is often misleading. However, the incidental observations at autopsy of incipient pulmonary tuberculosis and its relation to preexistent and apparently silent foci are of the greatest significance for the study of the pathogenesis of pulmonary tuberculosis. In the present review it is intended to discuss the different tuberculous lesions which are encountered incidentally at autopsy as the remnants of the primary or of the postprimary infection from the standpoint of their phthisiogenetic potentialities.

#### THE APICAL LESIONS

For practical purposes the apex of the lung may be defined as the portion of the upper lobe which is bordered inferiorly by the clavicle. The anatomic apex is the extrathoracic portion of the upper lobe, located above the first rib. This portion is also called the vertex pulmonis (Anders). Provided the thorax is normal in shape the geometrically highest point of the lung *in situ* (*culmen pulmonis*) is found intrathoracically just below the tuberculum of the first rib (Anders). The straight apical bronchus supplies an oblique, cone-shaped area the base of which faces upward.

The apex of the lung, in particular its highest point, the culmen, is the most favored site of postprimary tuberculous processes. The explanations for the disposition of the apex to tuberculosis are many. Thus, it is often referred to the compression of the apex by the first rib, to the poor respiratory ventilation and to the sluggish circulation of the blood in this area. According to Orsos the pulling of the diaphragm acts most intensively on the apex since it is reduced to a small area causing foci of relaxation on the surface of the apex. In these foci of relaxation foreign material is apt to be deposited.

Though the postprimary tuberculous lesions are most commonly encountered in the apex, it is also in the apex that these lesions show the greatest tendency to heal. The disposition of the apex to tuberculosis, therefore, does not mean that progressive pulmonary tuberculosis

tends to start in the apex. In recent years one has learned to distinguish several types of apical lesions which seem to differ as to their phthisiogenetic potentialities.

*Pleural Scars.*—These scars, which occupy the posterior aspect of the apex in the region of the culmen or extend over the vertex in the form of a cap, are from one to several millimeters thick. They are often very firm, of cartilage-like consistency, and their outer surface may be smooth and shiny or fixed to the thoracic wall by fibrous bands. Microscopically they consist of the thickened visceral pleura and a subpleural layer of indurated and sclerosed lung tissue with small round cell infiltrations and an occasional glandular tubulus. Since there are often no definite histologic evidences of their tuberculous origin, several authors (Lubarsch, Schuermann, Oberndorfer) have questioned whether one is justified in considering all these scars as tuberculous. I believe, however, with Schmincke, Graeff, Anders, Loeschke, Aschoff and Focke, that the great majority of them are the residues of abortive tuberculous infections. Wurm assumes that silicosis may lead to apical scars. However, the silicotic granulomas select chiefly the midportions of the lung. Nonspecific inflammations of the lung likewise do not gravitate to the apex. I do not have the impression that in influenza or pertussis (or measles) pneumonia the apexes of the lungs are particularly affected. Rubinstein emphasized that the purely fibrotic scars have no relation to progressing pulmonary tuberculosis.

*Discrete Miliary Tuberculosis of the Apex (Miliaris Discreta, Neumann).*—In young persons, especially in children below 15 years of age, one finds occasionally in the apex, just underneath the pleura, a group of pearly-gray nodules varying in size from that of a pinpoint to that of a pinhead, surrounded by normal or slightly congested lung tissue. In addition to these nodules the lung contains a well encapsulated or calcified primary lesion in some other area. On microscopic examination the nodules prove to be typical miliary tubercles chiefly of productive character (Huebschmann). There is a marked tendency to fibrotic involution, and the nodules become replaced by scar tissue, which under the influence of the emphysematous distention of the surrounding alveoli may be stretched so much that only a focal thickening of the stroma of the lung results. This focal interstitial fibrosis can be detected only by histologic examination. The small scars are of no significance as far as progression is concerned. In some instances, however, in which these scars are more marked, one may find a small bronchus or a bronchiolus the lumen of which contains a plug of inspissated caseous or caseocalcareous material, indicating that the lesions started to spread by aspiration. The dense scars may be visible on roentgen examination (*fibrosa densa*, Neumann).

The discrete miliary apical tuberculosis is undoubtedly hematogenous and may be described as an abortive early generalization restricted to the apexes (Huebschmann, Pagel, Neumann).

*Simon's Foci.*—Simon described in the lungs of children between 2 and 8 years of age symmetrical caseous nodules confined to the apexes. These nodules seem to develop immediately after the primary complex (subprimary) and are hematogenous, similar to the discrete apical miliary tuberculosis to which they are related (Huebschmann, Anders, Pagel). A characteristic feature of Simon's foci is their great tendency to calcification. In the early stages they are composed of confluent epithelioid cell tubercles with giant cells and central caseation. There is usually no perifocal exudation, and the bronchi are not involved (Anders). The tributary lymph nodes are found free from tuberculous changes. In spite of the limited activity which Simon's foci display, Pagel has expressed the opinion that in some instances they may lead to progression. Since the literature contains very few anatomic reports on Simon's foci, a personal observation may be described.

The case was that of a white girl, aged 5 years, who died of tuberculous leptomeningitis. At the medial aspect of the upper lobe of the right lung there was a well circumscribed, mortar-like primary lesion which measured 6 mm. in diameter. In the draining lymph nodes of that lobe calcified areas, some as large as 15 mm. in diameter, were found. Similar areas were present in the right epibronchial and paratracheal lymph nodes, while the lymph nodes at the left venous angle contained more recent caseous lesions. In the apex of each upper lobe an almost symmetrical, firm nodule was present. It measured 10:12:8 mm. in diameter and consisted of a dry caseous center surrounded by a thin, light gray capsule. About the nodule there were several yellow-gray areas varying up to 1 mm. in diameter which were embedded in moist and much congested lung tissue. The remaining portions of the lungs were free from changes.

*Aschoff-Puhl's Foci.*—Aschoff and his pupil Puhl have called attention to fibrocaseous nodules which occur in the apical and subapical parts of the upper pulmonary lobes and which increase in frequency with progressing age. The nodules are usually multiple and are considerably larger than the apical lesions described by Simon. Histologically one finds a caseous center surrounded by a thin, hyaline (Aschoff's specific) capsule and a thick, fibrotic (Aschoff's nonspecific) capsule. The latter is composed of atelectatic, indurated lung tissue rich in coal pigment and infiltrated by round cells. The caseous center may become calcified, but the calcification seldom is so marked that stony concretions are formed. Ossification is very rare. The tributary lymph nodes either are free from tuberculous changes or contain but a few abortive tubercles. According to Aschoff and Puhl, these histologic features are sufficiently characteristic to distinguish the apical nodules from the primary lesions, which have a thick specific and a thin

nonspecific capsule, tend to become ossified and are associated with marked tuberculous changes in the regional lymph nodes.

Aschoff is of the opinion that the nodules under discussion are due to an exogenous superinfection and are the most common source of the pulmonary phthisis. The majority of investigators agree with Aschoff that the apical nodules are postprimary, but with regard to the other points considerable disagreement of opinions exists. Pagel and Wurm have emphasized that the apical nodules are much too pleomorphic to be of uniform origin. Admitting that some of them are the result of an exogenous superinfection, they believe that the majority of them are hematogenous metastases. Aspiration from the primary lesion into the apex seems to be of little importance. The hematogenous infection of the apex may be part of an abortive early generalization in which the infection selects the disposed apical parts of the lung (Braeuning and Redeker). The Aschoff-Puhl foci then would be identical with the Simon foci, their larger size being due to an exacerbation of the originally small lesions. The hematogenous infection of the apexes may also occur in later life, e. g., from the exacerbation of a silent focus in a lymph node (Ghon's endoglandular exacerbation). Pagel's chief argument in favor of the hematogenous origin of the apical nodules is that he found them three times as often in generalized tuberculosis as in isolated pulmonary tuberculosis.

Birsch-Hirschfeld, Schmorl and Abrikosoff believed that the apical process started with a small tuberculous ulcer in the mucosa of a bronchus of the third to fifth order. Aschoff, Loeschke and Graeff stress the intra-alveolar and interalveolar location of the earliest lesion with secondary spreading to the bronchi. The earliest stage, apparently, has not yet been seen. Schuermann, who made a most careful, systematic study of the first postprimary tuberculous changes of the lungs, stated that even in the earliest stages which he was able to observe both the bronchi and the respiratory parenchyma were found involved. There are exudative caseous and acinous productive foci arranged about a bronchus the mucosa of which is transformed into caseous material.

As far as the exact location of the nodules is concerned, Anders stated that in over 85 per cent of the cases the lesions select the territory of the dorsal, apical bronchus, i. e., the culmen. The farther away from the culmen, the less frequently is the lung tissue affected. Loeschke, too, mentioned the region of the apical bronchus as the favorite site of the first postprimary tuberculous lesions of the lung. The ventral half of the apex is rarely affected (Zeiss). On the other hand, Schuermann summarized his findings as follows: About 60 per cent of the postprimary foci are located within from 3 to 4 cm. below the vertex. The rest of them are found at a lower level, sometimes

without any changes higher up. In general, the lesions at the lower level are larger and are related to larger bronchi than the lesions in the apex. They are also more apt to become transformed into small cavities. My own experience is fully in accord with that of Schuermann.

The apical caseous and caseocalcific nodules remain potentially active throughout life. They contain sometimes so many acid-fast bacilli that under low power magnification red areas are visible (Pagel). The progression may immediately follow the formation of the nodules, or the nodules may become well encapsulated and remain silent for a varying length of time, flaring up later. The continuous progression occurs chiefly in young persons. Schuermann gave the average age as 21.4 years. There are several routes for spreading. The infection creeps into adjacent alveoli and alveolar ducts, or the bacilli are carried by the blood and lymph stream into the surrounding lung tissue. The most important mode of progression, however, is by aspiration of infectious material from diseased bronchioli and bronchi into branches which come off from the same bronchus of the next higher order.

The exacerbation of foci at first silent is much more common than the direct continuation (Schuermann). It is a phenomenon of later life. In twenty-eight cases which Schuermann studied, the patients' ages varied between 24 and 76, with an average of 53. The indurative processes about the caseous nodules often lead to pleural retraction, to emphysematous distention of adjacent areas and to small bronchiectases (Pagel and others). The caseous or chalky centers of the nodules are apt to become liquefied or sequestrated. At autopsy one finds occasionally in the midst of the dense and anthracotic tissue a small cavity which contains a moderately firm calcium concretion or a pasty, light gray material. Microscopically the pasty material consists of fatty débris, cholesterol crystals and elastic fibers, and eventually also of calcium granules. Tubercl bacilli can be easily demonstrated by direct smears, culture or animal inoculation. After removing the content the cavity presents a smooth, light gray inner lining. If a sequestered or liquefied nodule breaks into one of the emphysematous blebs or bronchiectases previously described the way to aspiration and progression is opened. Some of the content of the small cavities may be expectorated, and after a severe attack of coughing the patient detects the peculiar looking material in his sputum. The demonstration of fragments of elastic fibers and of tubercle bacilli distinguishes this material from the nonspecific concretions which occasionally form in the lumens of bronchi.

Stefko attributes much significance to "alterative cavities" which may form about the nodules, leading to their sequestration. According to this author these parafocal or perifocal alterative cavities result

from a liquefaction of the lung tissue about the nodules which is due to a suddenly developing negative allergy, to a "breaking down of the symbiotic immunologic relation between host and micro-organisms." Another mode of exacerbation, finally, consists of the gradual transformation of the nonspecific capsule into tuberculous granulation tissue.

The causes which lead to the flaring up of the silent lesions are difficult to determine. Stefko's explanation is purely theoretical. Autopsy observations show that this flaring up is most commonly encountered in patients with diabetes mellitus, in pregnant women and in people who died of chronic wasting diseases, especially malignant tumors. Exogenous superinfection may stimulate the old foci to new activity by means of a tuberculin-like action of the newly inhaled tubercle bacilli on the preexistent lesion. Nontuberculous changes about the encapsulated nodules may favor their progression. This is illustrated by the following case:

A white woman, 41 years of age, died of an ulcerated carcinoma of the rectum with extensive metastases to the liver. In the upper third of the upper lobe of the right lung there was a single metastasis which measured 16 mm. in diameter and compressed the proximal portion of the subapical bronchus. Distal to the compression the bronchus was dilated and filled with thick, creamy pus. The lining of the bronchus was transformed into caseous material. The dilated bronchus terminated in a subpleural fibrocaseous nodule which measured 15 mm. in diameter and was partially liquefied. About the apical, subapical and horizontal bronchi there were multiple small foci of exudative caseation. I believe that in this case the exacerbation and acinous spreading of the subapical lesions were stimulated by the compression of the subapical bronchus by the metastasis. The tumor cachexia may have contributed to the exacerbation, but a nodule in the apex of the upper lobe of the left lung showed no signs of activity.

A critical analysis of the potentialities of the apical lesions on the basis of pathologico-anatomic observations leads to the conclusion that the caseous and caseocalcific nodules may cause dissemination. This dissemination, however, is usually limited and of small caliber. Are there any indications that the apical lesions may indirectly become the source of progressive phthisis by way of the infraclavicular infiltration?

#### THE INFRACLAVICULAR INFILTRATIONS

In the literature on tuberculosis the term infiltration is usually applied to lesions which are marked by their lability. These infiltrations may form and disappear quickly and may leave no roentgenologically demonstrable residue. On the other hand, they often show rapid caseation, liquefaction and ulceration followed by massive bronchogenic spreading. Though the infiltrations show a predilection for the infraclavicular region they may affect any portion of the lung. They are not specific of a certain stage of the tuberculous disease in the sense of

Ranke, since they may develop secondarily about primary lesions, about their lymph node component, about lesions of the generalization period and in incipient, isolated, progressive pulmonary tuberculosis. Some cases of epituberculosis are apparently due to an extensive perifocal infiltration about caseous foci in the hilus lymph nodes.

About the pathology of the transient infiltrations and the earliest stages of the caseous and ulcerative infiltrations little is known. There are very few reports (Assmann, Elias, Kudlich and Reimann, Loeschke, Mischkowsky, Pagel, Rubinstein, Schmincke, Schuermann), and the majority of these reports deal with rather advanced changes. It is likely that the majority of the transient infiltrations are purely exudative processes which do not reach the stage of caseation. Pagel spoke of a viscid, plasmatic exudate in the alveoli, rich in mononuclear alveolar phagocytes. The alveolar septums are thickened and infiltrated and contain an occasional giant cell or abortive miliary epithelioid cell tubercle. Thus, the picture resembles a gelatinous pneumonia. In other instances, circulatory processes in the form of prestasis, stasis and hemorrhage predominate (Schuermann). These lesions are capable of complete resorption. Delay of the resorption may be due to fibrosis of the alveolar septums, collapse induration and nonspecific organization of the alveolar exudate.

Some of the transient infiltrations center about small caseous nuclei (Assmann). They are then perifocal reactions in a highly sensitive person (Ranke, Tendeloo). After resorption of the perifocal exudate the remaining caseous area may be too small to be visible on x-ray examination. The caseous area may become gradually replaced by connective tissue and the emphysematous distention of the surrounding alveoli may stretch the scar, making it fit into the framework of the adjacent lung parenchyma. There is then an almost complete restitutio ad integrum.

When the infiltrations take a turn for the worse the exudation is followed by caseation. The caseous material becomes liquefied; it finds its way into a bronchus, and a cavity is formed. The cavities which develop from the liquefied infiltrations are characteristic. They are usually spherical and possess a thin wall the inner lining of which is smooth. After the perifocal reaction has subsided the cavities are surrounded by practically normal lung tissue. These "punched-out" cavities differ in many respects from the cavities of chronic ulcerative tuberculosis, which are surrounded by a thick, sclerotic wall the inside of which is covered by a layer of caseous material. The rapid liquefaction of the caseated infiltration is very dangerous, since it opens blood vessels and bronchi. Hemorrhage and aspiration result, and the infection spreads rapidly to other portions of the lung.

The infraclavicular infiltrations do not lead to specific changes in the regional lymph nodes. If tuberculous changes are present, they are, as a rule, older than the process in the lung (Pagel, Schmincke, Schuermann, Loeschke, Beitzke).

The different pathologic-anatomic manifestations of the tuberculous infection have often been linked to immunity and allergy and Ranke especially suggested a geneal classification of tuberculosis on the basis of immunologic and allergic phenomena. Ranke's conception has often been interpreted wrongly, and there is so much discrepancy in the modern literature that it has become somewhat discredited to speak of immunity and allergy in tuberculosis. The critical observer, however, cannot help feeling that the infraclavicular infiltrations, with their massive exudation and quick liquefaction, carry the earmarks of an allergic, in particular hyperergic, inflammation. They are the response of a sensitized organism. In this connection, it may be recalled that allergy and immunity are not related (Rich) and that hyperergic inflammation does not indicate increased protection.

#### THE RELATIONS BETWEEN THE INFRACLAVICULAR INFILTRATIONS AND THE APICAL LESIONS

Loeschke and Huebschmann maintain that the infraclavicular infiltrations are always secondary to apical lesions. A similar opinion is held by Aschoff and Graeff. In the preceding section I described a caseous inflammation of the bronchioli and finest bronchi which is found in connection with the apical caseous nodules. Aspiration of infectious material from the finest bronchi may cause the tuberculosis to spread to other branches of the dorsal apical bronchus, and minute aspiration foci are formed, which are chiefly productive. These minute foci have a tendency to heal, but the process may slowly spread in the cranio-caudal direction (small shotlike dissemination of Loeschke).

By direct continuation or following an exacerbation the caseous endobronchitis may descend into larger branches until it reaches the main apical bronchus, the lumen of which becomes filled by a cast of caseous material. Loeschke stated that the infraclavicular infiltrations result from the massive aspiration of tuberculous material into the subapical and horizontal bronchi. The massive aspiration causes exudative inflammation with caseation and ulceration (coarse shotlike dissemination). Loeschke's interpretation has been confirmed by Schuermann, Pagel and Aschoff, and I have repeatedly been able to trace an infraclavicular caseous or ulcerative process to an older focus in the apex. Loeschke's explanation, however, does not take into account the transient infiltrations, which, though extensive, may become completely resorbed. Schuermann stressed the fact that coarse shotlike dissemination is not

always bound to the caseous inflammation of a larger bronchus. It may also follow small initial lesions when many bacteria of great virulence are aspirated.

In addition to the aspiration from the apex into the infraclavicular region there occur infraclavicular infiltrations which are not related to apical lesions. In these cases, either the apex contains only silent and completely encapsulated nodules or fibrotic scars or it is free from changes and the infraclavicular infiltration is the first manifestation of the postprimary pulmonary tuberculosis (Elias, Kudlich and Reimann, Mischkowsky, Pagel, Schuermann, and others).

The acute formation of an infraclavicular cavity without apical lesions is illustrated by the following case:

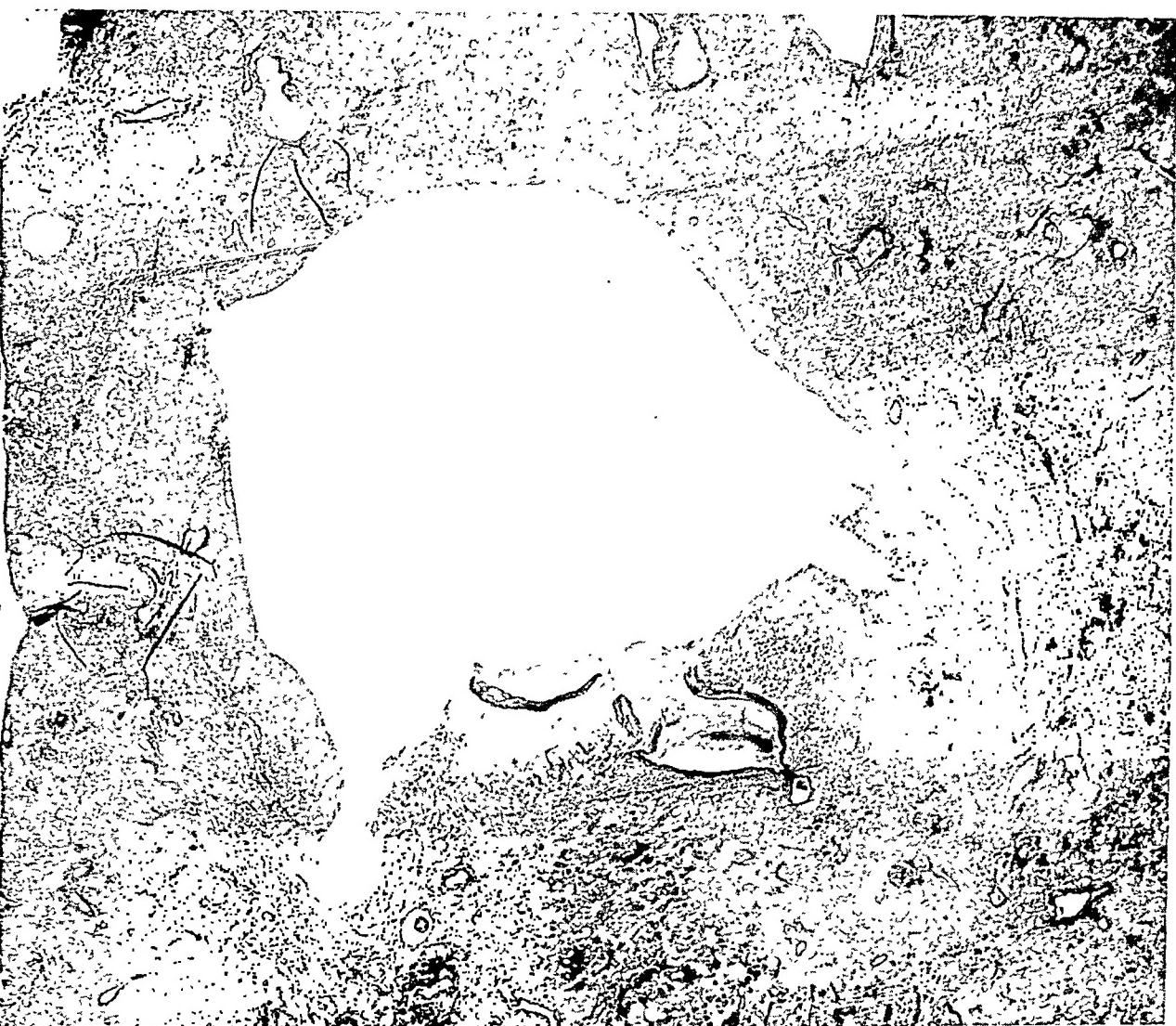
A colored man, aged 38, stated that he was perfectly well until five weeks before his admission to the hospital, when he experienced a sudden chill with severe headache which was followed by fever and cough productive of a mucoid material. During the last three weeks prior to admission he lost 5 pounds (2.3 Kg.) in weight and was in bed most of the time. There was no history of tuberculosis in his family. On admission his temperature was 102.4 F. The respiratory rate was 28, and the pulse rate, 112. There was diminished resonance over the entire left lung and an area of dulness was noted between the left clavicle and the nipple. Over this area amphoric bronchial breathing and bubbling râles were heard. Roentgen examination revealed clear apexes and an opacity in the middle third of the left lung. The sputum contained many tubercle bacilli. The blood count showed hemoglobin, 80 per cent; erythrocytes, 4,200,000, and leukocytes, 4,400. After a stay of a few days in the hospital the patient suddenly became very dyspneic and died.

At autopsy the left lung was distended and heavy, and multiple small areas of consolidation were felt in the lower two thirds of the upper lobe and in the upper third of the lower lobe. At the border between the upper and the middle third of the upper lobe there was a sharply circumscribed, roughly spherical cavity which measured 23 mm. in diameter and was filled with blood. The cavity was lined by a friable, light yellow-gray membrane and was surrounded by confluent, opaque, light yellow areas which averaged 10 mm. in diameter. These areas extended throughout the lower half of the upper lobe and the upper half of the lower lobe. The lung tissue about the areas was deep purple-red and very moist. The bronchi contained bloody mucoid material. The lymph nodes at the hilus were small, firm and anthracotic. The right lung contained air, and there were small areas of aspirated blood in the lower lobe. The apexes of both lungs were free from scars or fibrocaseous nodules. Near the base of the lower lobe of the right lung there was a subpleural calcified nodule the size of a cherry stone.

On microscopic examination no tuberculous lesions or scars could be detected in the apexes. The cavity was lined by a layer of caseous material which contained an enormous number of acid-fast bacilli. The caseous material rested on a very thin membrane which was densely infiltrated by small round cells and contained only a few Van Gieson red fibrils. There were many congested capillaries. The cavity communicated with two small bronchi the mucosa of which had been replaced by caseous material. The larger of the two bronchi had a diameter of 4 mm., and

the accompanying artery opened into the cavity (see illustration). About the cavity there were many foci of a caseating exudation with numerous tubercle bacilli and an intense perifocal reaction.

While in this case the cavity was in an early stage of formation the next observation is an example of a typical "punched-out" cavity.



An early infraclavicular cavity. Note the eroded artery in the lower wall of the cavity and the extension of the caseation into the adjacent bronchus, the marked perifocal reaction about the cavity and the foci of exudative caseation. Weigert's elastin stain; magnification,  $\times 5$ .

The patient, a white woman 45 years of age, was brought to the hospital in coma and died four hours later. The autopsy revealed chronic meningo-encephalitis with severe acute degenerative changes of the ganglion cells. On the lateral aspect of the upper lobe of the left lung, at the border between the upper and the middle third, there was a firm subpleural area from 2 to 3 cm. in diameter. Sectioning

this area exposed several dry, caseous foci from 1 to 6 mm. in diameter which were located posteriorly to a spherical cavity 18 mm. in diameter. The cavity was lined by a thin, light purple-gray membrane the inside of which appeared smooth. The bronchi of this region were dilated and filled with a mucopurulent material. The apex was adherent to the wall of the chest and contained a small anthracotic scar. The apical bronchus was free from changes, as were also its tributaries. The hilus lymph nodes were small and deeply anthracotic. Microscopically the dry areas proved to be foci of caseous bronchopneumonia with a moderate perifocal reaction. The lining of the cavity was formed by Van Gieson's red connective tissue. There was no caseous material on the inside of this membrane, and the lung tissue about the cavity was not indurated.

In some instances the apex and the subapical region are found diffusely involved and the process is so uniform in character that one is justified in assuming a simultaneous involvement of both parts. Even serial sections fail to disclose an old focus. A case of this type may be briefly described.

A white man, aged 49, died as a result of fatty necrosis of the pancreas with diffuse serofibrinous peritonitis. The right lung was covered by a loosely adherent, soft membrane of fibrin. The upper fourth of the upper lobe felt firm, while the remaining portions were crepitant. On the sectioned surface the firm area was uniformly granular, dry and pale yellow. The apical and subapical bronchi were completely obscured by this area, as was also the distal part of the horizontal bronchus. In the lower portion of the upper lobe of the right lung and in the posterior part of the lower lobe of each lung there were several dry, yellow-gray areas, varying in size up to that of a pinhead. On the anterior aspect of the upper lobe of the left lung, near the lower border, there was a fibrocalcareous primary lesion which measured 10 mm. in diameter. The lymph nodes at the hilus of each lung were small and contained a few fibrocalcareous nodules. Microscopic examination of the consolidated area of the upper lobe of the right lung showed diffuse caseation of the alveolar walls and alveolar exudate with preservation of the elastic fibers. There were no epithelioid cell tubercles or giant cells nor any older changes.

Finally, it may be mentioned that apical lesions may be caused by aspiration from the infraclavicular process.

#### THE PATHOGENESIS OF THE INFRACLAVICULAR INFILTRATIONS

In a recent publication, Terplan reported that he had found in Buffalo a relatively small percentage of primary lesions in children and young adults. After the age of 20 years the pulmonary primary lesions increased in frequency. The author discusses the possibility that primary infection may also occur after the second and third decades of life. My experience in Chicago has been similar to that of Terplan, namely, after the age of 30 years residues of the primary infection are more common than below this age. Kalbfleisch observed typical recent pulmonary primary lesions in old persons and assumed that in these instances the effect of the primary infection acquired in early life had

completely subsided. The question will therefore arise whether a primary lesion with a marked perifocal reaction may present itself under the picture of an early infraclavicular infiltration. There are, however, principal differences between a primary lesion and an infraclavicular infiltration. The primary lesion is characterized by its stability and by its tendency to become encapsulated. The infraclavicular infiltration is labile. It may clear up completely or may rapidly progress to ulceration and cavitation. The primary lesion is followed by marked caseous tuberculosis of the tributary lymph nodes, while the infraclavicular infiltration does not spread to the lymph nodes. It is, however, possible that a primary lesion may cause an infraclavicular infiltration by aspiration. Mischkowsky described the case of a girl, aged 19, in which autopsy revealed a caseous and ulcerated infiltration in the infraclavicular region of the right lung which he thought was due to a bronchogenic dissemination from an exacerbating primary lesion in the upper lobe of the left lung.

When Assmann published his first observations on infraclavicular infiltrations he came to the conclusion that they were the result of a postprimary exogenous superinfection. Many authors have accepted Assmann's point of view, stressing that the infiltrations are most commonly found in people known to have been exposed, such as physicians, nurses and autopsy helpers. It is, however, likely that these people are more apt to come under early observation than persons with other occupations. Kudlich and Reimann came to the conclusion that in their case exogenous superinfection offered the best explanation. Their explanation has not been accepted by Pagel. In the case of early infraclavicular cavitation which I have described here there were no evidences of an exacerbating pulmonary or extrapulmonary focus which may have served as a source of the recent process. Pagel, too, has admitted that some of the infraclavicular infiltrations may result from a new, exogenous infection with tubercle bacilli (see also Loeschke, Schuermann, Aschoff and Graeff).

Adler, Albert, Braeuning and Redeker, Lydtin, Ulrich and others have offered clinical and roentgenologic evidences that hematogenous foci may progress to form early infraclavicular infiltrations and pulmonary phthisis. Pagel emphasized that in generalized forms of tuberculosis which are due to a prolonged dissemination of tubercle bacilli through the blood stream punched-out cavities in the infraclavicular region are encountered which are identical with the cavities that develop from early infiltrations (see also Schuermann and Pinner). In generalized tuberculosis the lesions tend to become stabilized, and massive bronchogenic spreading is relatively rare. There is a certain antagonism between generalization and local progression. In some instances,

however, the metastases are confined to the lungs and select the infra-clavicular region. At first the hematogenous metastases become encapsulated and even calcified. As in the case of the apical lesions previously described, exogenous superinfection or decrease in resistance may cause a flaring up and progression of the isolated infraclavicular foci. The source of the hematogenous foci can be found in an active process in the lymph nodes or in tuberculosis of the bones, joints, genito-urinary tract, etc.

Liebermeister spoke of tuberculous hemorrhagic infarcts of the lung which may give a roentgenologic picture similar to that of an infra-clavicular infiltration. In one of his cases he described a wedge-shaped hemorrhagic area which extended from the hilus through the lower part of the upper lobe. At the hilus a cherry-sized caseated lymph node was present, and there was severe tuberculous involvement of the blood vessels. Similar lesions were mentioned by Graeff and Küpferle and by Tendeloo. In this connection the "round tuberculous foci" may be discussed.

*The Round Tuberculous Foci.*—Albert has called attention to round tuberculous foci of the lung which are usually discovered incidentally on roentgen examination of the chest. These foci do not cause any symptoms. The patient is afebrile, the sputum is free from tubercle bacilli and the sedimentation rate of the erythrocytes is not increased. Because they are so sharply defined the round foci resemble metastases of a tumor. They have also been confused with cysticercosis (Jacksch von Wartenhorst). The foci are single or multiple, and as many as fourteen have been counted in a single case. They occur in any part of the lung but are most common in the infraclavicular region.

The round tuberculous foci often remain stationary for many months or even for several years. This inactivity distinguishes them from the infraclavicular infiltrations. Their ultimate fate varies. Bruck observed multiple foci in the lung of a woman, 32 years of age, which disappeared spontaneously. Some of the foci become calcified, while others are gradually replaced by connective tissue and transformed into scars. Albert and Straub emphasized that the prognosis is not always favorable. After a period of inactivity the foci may start to progress or they may break down and form a cavity with the danger of aspiration. Their relation to the infraclavicular infiltrations is still under discussion. Pagel, among others, believes that they may become the starting point of pulmonary phthisis. The symmetrical location in both lungs suggested to Albert a hematogenous origin.

Anatomically the round foci are purely exudative and caseous (Albert, Lachmann). The lymph nodes are usually not involved. Anders observed a patient with caseation in the regional lymph nodes

and considered the lesion as due to a primary infection. Lachmann isolated bovine tubercle bacilli, while Albert obtained a strain of human tubercle bacilli. Since very few anatomic reports have been published, a typical case may be described. Unfortunately, no roentgen picture was made during the patient's brief stay in the hospital.

A white woman, 36 years of age, died after a stay in the hospital of twenty hours. The clinical picture was dominated by severe vaginal bleeding which led to a provisional diagnosis of incomplete abortion. At autopsy a fibrocaseous primary lesion 10 mm. in diameter was found in the middle lobe of the right lung. The lymph nodes at the hilus of the right lung contained several dry, cheesy nodules which were surrounded by a thick fibrous capsule. In some of the nodules the capsule was partially destroyed, and groups of confluent tubercles with central caseation were found about the older lesions. The paratracheal, bifurcation, peri-biliary, peripancreatic and periaortic lymph nodes were the sites of numerous, confluent, soft, caseous areas which varied from 25 to 40 mm. in diameter. There was a bilateral caseosuppurative salpingitis, as well as a discrete miliary dissemination to the spleen, liver and kidneys and a terminal endocarditis of the mitral valve. The endometrium was pale and smooth.

In the subapical portion of the upper lobe of the left lung there were two sharply circumscribed flat, round nodes, 15 and 17 mm. in diameter. They were firm and were composed of a homogeneous light yellow-gray material. In the subapical portion of the upper lobe of the right lung a single similar node 18 mm. in diameter was found. Histologic examination of the nodes showed diffuse caseous pneumonia. In sections stained for elastin the elastic fibers of the alveoli, bronchi and blood vessels were seen to be well preserved. About the caseous area there was a narrow zone in which the alveoli contained a fibrinocellular exudate. Tubercle bacilli could not be demonstrated.

I believe that this case proves the hematogenous origin of the round foci. The source of the hematogenous dissemination was found in the progressive tuberculosis of the lymph nodes which followed the endoglandular exacerbation of older lesions in the hilar lymph nodes. The foci in the lungs and those of the tuberculous salpingitis were of about the same age. In addition to these older hematogenous seedlings there was a recent miliary dissemination.

*Summary.*—The present knowledge of the pathology and pathogenesis of the infraclavicular infiltrations can be summarized as follows: Pathologico-anatomic observations confirm the clinical conception of the great significance of acute exudative infraclavicular processes as the source of progressive pulmonary tuberculosis. In the majority of the cases these infraclavicular processes reveal relations to older apical lesions. Thus they form the bridge between apical tuberculosis and pulmonary phthisis. In some instances, the infraclavicular infiltrations develop independently of or without apical foci. Clinicians give the incidence of the apical beginning of pulmonary tuberculosis as from 2.6 to 7.6 per cent (Braeuning and Redeker, Lydtin, Kayser-Petersen, Edel and Adler, Rubinstein). From the standpoint of the morbid

anatomist these figures are undoubtedly too low. Because of the scarcity of suitable material exact pathologic data are not yet available. Hematogenous infection of the lung is apt to produce the clinical and anatomic picture of infraclavicular infiltration and cavitation. There are, however, a considerable number of cases which strongly suggest that infraclavicular infiltrations may also develop from exogenous superinfection.

#### THE PRIMARY COMPLEX

*The Pulmonary Primary Lesion.*—The relation between the pulmonary primary lesions and the infraclavicular infiltrations has been discussed in the preceding section. In general, the primary lesions do not play an important rôle in causing isolated progressive pulmonary tuberculosis. Blumenberg assumes that in adults the primary infection of the lung does not take the typical course with encapsulation and caseation of the regional lymph nodes but rather tends to direct progression. Pagel and Beitzke do not agree with Blumenberg, and according to Pagel, Schuermann, Kalbfleisch, Terplan and many others the primary lesions contracted after adolescence are identical with those acquired in early life.

Exacerbation of an encapsulated primary lesion of the lung has often been described. In the literature reference is made to transient infiltrations about primary lesions, to extracapsular tubercle formation and to ulceration and sequestration of primary lesions (Pagel, Beitzke, Ghon, Schmincke and others). In a man 24 years of age who had committed suicide Wurm found a calcified primary lesion of the lung which was surrounded by recent tuberculous changes. The same author described, in a boy, 14 years of age, an ulcerated primary lesion of the lung (Schmincke's primary cavity type b) with massive bronchogenic dissemination. The majority of the exacerbating primary lesions, however, do not progress. The transient infiltrations are resorbed without leaving any traces and the extracapsular tubercles become fibrosed. Steffko referred to alterative cavities about the primary lesions which cause sequestration of the lesions. These sequestered primary lesions are usually expectorated.

Siegen described a vascular pedicle through which the primary lesion is connected with the surrounding lung tissue. This pedicle offers a pathway to the tubercle bacilli in the center of the lesion. The significance of the vascular pedicle has been questioned by Pagel. It has not been definitely established how long viable tubercle bacilli persist in calcified and petrified primary lesions. At present the majority of investigators believe that the infectiousness of the primary lesions decreases markedly with progressing age. Sweany made the statement that he had not been able to decide whether the regressive changes

which cause the destruction of the wall of the primary lesions may lead to a liberation of latent bacilli. He considers it possible that nature may defeat her own purpose by opening the way to progression of the infection. I believe that the ossification which so often takes place in older primary lesions indicates absence of tubercle bacilli from the calcified center. This ossification is initiated by the ingrowth of young granulation tissue which is derived from the capsule. If viable tubercle bacilli were present in the calcified material which is destroyed and replaced by the granulation tissue, tubercles would be formed as a specific reaction to the surviving bacilli. In a large series of primary lesions in various stages of ossification I have not encountered a single instance of tubercle formation by the young granulation tissue. I believe that the persistence of tubercle bacilli in the Aschoff-Puhl foci interferes with the ossification of these lesions.

*The Lymph Node Component.*—In the lymph node component of the primary lesions exacerbation is undoubtedly more common than in the primary lesion itself. The significance of this endoglandular exacerbation for the hematogenous infection of the lung has already been referred to. The majority of authors assume that retrograde lymphatic spreading from an exacerbated focus in a hilar lymph node into the lung is rare. Wurm observed the perforation of an exacerbated lymph node tubercle into a large bronchus with aspiration.

#### THE REMNANTS OF EARLY GENERALIZATION

The primary tuberculous infection is apt to lead to an invasion of the blood stream. In the severe form this early generalization causes the acute miliary tuberculosis which immediately follows the primary complex and shows a high incidence of involvement of the meninges. In the great majority of the cases sufficient resistance has developed during the formation of the primary complex to check the invasion of the blood stream, and no colonization or an abortive colonization takes place in the different organs. This abortive generalization escapes clinical detection. The small hyaline or calcified nodules which are sometimes found at autopsy in the spleen, lungs, liver, kidneys and other organs are the remnants of the checked early generalization. It is from the isolated hematogenous foci that progressive tuberculosis of the bones, joints, kidneys, tubes, etc., takes its origin (Ranke). As far as the lungs are concerned the hematogenous foci may be confined to the apices (Simon's foci and perhaps also some of the Aschoff-Puhl foci). They may also be found scattered throughout the lungs, and if they are of sufficient size and densely calcified, they may be visible on roentgen examination and may be mistaken for multiple primary lesions.

Schmincke expressed the opinion that any of the pulmonary nodules may flare up and lead to progressive tuberculosis. Wurm, a pupil of Schmincke, compared the frequency of the calcified nodules in tuberculous and nontuberculous lungs and found a much higher incidence in the former. There were also microscopic evidences of exacerbation and progression.

#### BIBLIOGRAPHY

- Abrikosoff: *Virchows Arch. f. path. Anat.* **178**:173, 1904.  
 Adler, H.: *Beitr. z. Klin. d. Tuberk.* **80**:22, 1932.  
 Albert, A.: *Beitr. z. Klin. d. Tuberk.* **78**:647, 1931.  
 Anders, H. E.: *Verhandl. d. deutsch. path. Gesellsch.* **24**:186, 1929; *Med. Klin.* **29**:777, 1933.  
 Aschoff, L.: *Klin. Wchnschr.* **8**:1, 1929.  
 Ueber die natuerlichen Heilungsvorgaenge bei der Lungenphthise, Munich, J. F. Bergmann, 1921.  
 Assmann, H.: *Beitr. z. klin. d. Tuberk.* **60**:526, 1923; *Med. Klin.* **28**:1335, 1932.  
 von Baumgarten, P.: *Beitr. z. path. Anat. u. z. allg. Path.* **69**:27, 1921.  
 Beitzke, H.: *Beitr. z. Klin. d. Tuberk.* **65**:291, 1926.  
 Birsch-Hirschfeld, F. H.: *Deutsches Arch. f. klin. Med.* **64**:58, 1894.  
 Blumenberg, W.: *Klin. Wchnschr.* **8**:505, 1929.  
 Braeuning, H., and Redeker, F.: Studien zur Entwicklung der menschlichen Lungenphthise: I. Die hämatogene Lungentuberkulose des Erwachsenen, Leipzig, Johann Ambrosius Barth, 1931.  
 Bruck, S.: *Am. J. Roentgenol.* **31**:319, 1934.  
 Elias, B.: *Beitr. z. Klin. d. Tuberk.* **80**:175, 1932.  
 Fishberg, M.: Pulmonary Tuberculosis, ed. 4, Philadelphia, Lea & Febiger, 1932.  
 Focke: *Beitr. z. Klin. d. Tuberk.* **59**:228, 1924.  
 Gekler: *Arch. Int. Med.* **20**:32, 1917.  
 Ghon, A.: *Ztschr. f. d. ges. exper. Med.* **50**:25, 1926.  
 —and Kudlich, H.: Die Eintrittspforten der Infektion vom Standpunkt der pathologischen Anatomie, in Engel, S., and von Pirquet, C.: *Handbuch der Kindertuberkulose*, Leipzig, Georg Thieme, 1930.  
 Graeff, S.: *Beitr. z. Klin. d. Tuberk.* **70**:173, 1928.  
 —and Küpferle, L.: Die Lungenphthise, Berlin, Julius Springer, 1923.  
 Huebschmann, P.: Pathologische Anatomie der Lungentuberkulose, Berlin, Julius Springer, 1928.  
 Kalbfleisch, H. H.: *Ergebn. d. ges. Tuberk.* **4**:47, 1932.  
 Kudlich, H., and Reimann, F.: *Ztschr. f. Tuberk.* **55**:289, 1930.  
 Lachmann, E.: *Fortschr. a. d. Geb. d. Röntgenstrahlen* **43**:407, 1931.  
 Liebermeister, G.: *Ztschr. f. Tuberk.* **62**:83, 1931.  
 Loeschcke, H.: *Beitr. z. Klin. d. Tuberk.* **68**:251, 1928; **81**:171, 1932.  
 Lubarsch, O.: *Virchows Arch. f. path. Anat.* **213**:417, 1913.  
 Lydtin, K.: *Beitr. z. Klin. d. Tuberk.* **67**:235, 1927; *Ztschr. f. Tuberk.* **49**:1, 1928; **57**:153, 1930.  
 Mischkowsky, R.: *Ztschr. f. Tuberk.* **67**:17, 1933.  
 Neumann, W.: *Beitr. z. Klin. d. Tuberk.* **40**:1, 1919.  
 Oberndorfer, quoted by Pagel.  
 Orsós, F.: *Beitr. z. Klin. d. Tuberk.* **70**:504, 1928.

- Pagel, W.: Deutsche med. Wchnschr. **57**:2094, 1931; Ztschr. f. Tuberk. **65**:197, 1932; Ergebn. d. ges. Tuberk. **5**:231, 1933.
- Lungentuberkulose, in Henke, F., and Lubarsch, O.: Handbuch der speziellen pathologischen Anatomie und Histologie, Berlin, Julius Springer, 1930, vol. 3, pt. 2, p. 139.
- Pinner, M.: Am. J. Roentgenol. **31**:442, 1934.
- Puhl, H.: Beitr. z. Klin. d. Tuberk. **52**:116, 1922.
- Ranke, K. E.: Ausgewählte Schriften zur Tuberkulosepathologie, Berlin, Julius Springer, 1928.
- Rich, A. R.: Bull. Johns Hopkins Hosp. **47**:181, 1930.
- Rubinstein, H.: Beitr. z. Klin. d. Tuberk. **70**:773, 1928. Spaltenprozesse, Leipzig, Johann Ambrosius Barth, 1934.
- Schmincke, H.: Ztschr. f. ärztl. Fortbild. **26**:105, 1929.
- Schmorl, G.: München. med. Wchnschr. **48**:1995, 1901; **49**:1379, 1902.
- Schuermann, P.: Beitr. z. path. Anat. u. z. allg. Path. **83**:551, 1930; Schweiz. med. Wchnschr. **63**:1145, 1933.
- Siegen, H.: Beitr. z. Klin. d. Tuberk. **63**:143, 1926.
- Simon, G.: Beitr. z. Klin. d. Tuberk. **67**:467, 1927; **81**:194, 1932.
- Stefko, W. H.: Beitr. z. Klin. d. Tuberk. **82**:566, 1933.
- Straub, H.: Ztschr. f. klin. Med. **121**:515, 1932.
- Sweany, H. C.: Am. Rev. Tuberc. **27**:559, 1933.
- Cook, C. E., and Kegerreis, R.: Am. Rev. Tuberc. **24**:558, 1931.
- Tendeloo, N. P.: Krankheitsforschung **6**:159, 1928; Beitr. z. Klin. d. Tuberk. **6**:329, 1906.
- Terplan, K.: Am. Rev. Tuberc. **29**:77, 1934.
- Ulrici, H.: Beitr. z. Klin. d. Tuberk. **77**:267, 1931; **81**:183, 1932.
- Wessler, quoted by Fishberg.
- Wurm, H.: Beitr. z. path. Anat. u. z. allg. Path. **75**:399, 1925; **79**:209, 1927; Beitr. z. Klin. d. Tuberk. **81**:707, 1932; Klin. Wchnschr. **13**:41, 1934.
- Zeiss, B.: Beitr. z. Klin. d. Tuberk. **64**:463, 1926.

## News and Notes

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**University News, Promotions, Resignations, Appointments, Deaths, etc.**—Herman A. Heise has been appointed director of the laboratories in the Columbia Hospital and the Children's Hospital in Milwaukee.

Otto Folin, professor of biologic chemistry in the Harvard University Medical School, Boston, died on October 26, at the age of 67.

George R. Minot and William P. Murphy of Harvard University Medical School, Boston, and George H. Whipple of the University of Rochester, New York, have been awarded the Nobel Prize, 1934, "for liver treatment in anemia."

Kornel L. Terplan, research professor of pathology in the University of Buffalo School of Medicine, has been appointed head of the department of pathology and bacteriology to take the place of Herbert U. Williams, retired.

Santiago Ramón y Cajal, the great Spanish neurologist and histologist, died on October 18, at the age of 83.

Carlos Chagas, director of the Oswaldo Cruz Bacteriological Institute in Rio de Janeiro, died on November 8, at the age of 55. He described the form of trypanosomiasis of the thyroid which is found in the interior of Brazil and known generally as Chagas' disease.

Carl L. Spohr has been made acting head of the department of pathology in the Ohio State University College of Medicine, Columbus, taking the place of the late Ernest Scott.

John A. Kolmer has resigned as professor of medicine in Temple University.

A department of bacteriology has been established in the University of Southern California, Los Angeles, under the direction of Carl C. Lindegren, recently assistant in microbiology at the Mellon Institute, Pittsburgh.

Edwin O. Jordan, formerly chairman of the department of hygiene and bacteriology in the University of Chicago, was awarded the Sedgwick Memorial Medal for distinguished service in public health at the meeting of the American Public Health Association in Pasadena last September.

**Society News.**—The Society of American Bacteriologists will hold its thirty-sixth annual meeting in Chicago, on December 27 to 29, 1934.

The Fifteenth International Physiological Congress will be held in Leningrad and Moscow from August 9 to 17, 1935, under the presidency of Ivan P. Pavlov.

**Resolution in Respect to Radiodermatitis.**—The following resolution was adopted by the American Radium Society at the Cleveland Session on June 12, 1934:

"WHEREAS, It has been proven that radium and x-rays, when used properly, and in sufficient quantity, are efficient in the treatment of cancer in certain locations, and

"WHEREAS, There is a general fear in the public mind from x-ray or radium burns, which because of this fear, prevents competent radiologists from using sufficient radium or x-ray to produce the best results.

*"Be It Resolved,* That we as radiologists recognize that in the treatment of malignant disease, it is often necessary to carry the treatment on to the extent of producing a violent reaction in the surrounding tissues, which may cause the skin to peel, and blisters to form, in order to give sufficient treatment to overcome the malignant disease. We believe, therefore, that it is justifiable to produce a second degree radiodermatitis when necessary.

This resolution has been approved by the Section on Radiology of the American Medical Association and by the American College of Radiology.

# Abstracts from Current Literature

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## Experimental Pathology and Pathologic Physiology

THE FLOW AND COMPOSITION OF LYMPH IN RELATION TO EDEMA. A. A. WEECH,  
E. GOETTSCH AND E. B. REEVES, *J. Exper. Med.* **60**:63, 1934.

The capacity of the lymphatics for removing fluid from the tissues greatly exceeds the rate at which freshly formed tissue fluid can be made available for removal. Edematous regions can be rendered nonedematous by the application of measures which activate the lymphatics, such as massage, passive motion or normal exercise. During continuous activity the rate of lymph flow is at first variable and later relatively constant. The constant rates of flow must correspond to the production of fresh lymph. A study of the constant rates indicates that the formation of lymph in edema is certainly only slightly greater, and possibly not greater, than under conditions of normality. When the protein of the plasma decreases, the protein of the lymph is also lowered. Loss of protein takes place at a faster rate from the lymph than from the plasma, so that the serum protein-lymph protein quotient is greater for the edematous than for the normal animal. In edematous animals the concentration of protein in the lymph is of the same order of magnitude as the concentration in the edema fluids. The two fluids are not, however, identical in composition. Minor fluctuations in the protein content of the lymph always occur during a period of continuous collection. The factors involved in the circulation and accumulation of tissue fluid are discussed. Reasons are given for offering the following suggestions: Significant differences in tissue pressure or tension exist between the states resulting from quiescence and activation of the lymphatics. The differences give rise to variations in the relative areas of capillary wall functioning for filtration, and reabsorption may be completely in abeyance. A decline in the proteins of the plasma may be associated with a diminished permeability of the capillaries. Such a lowering of capillary permeability would account for two features, both of which have been demonstrated: (1) failure to observe an appreciable increase in the rate of lymph formation in the edematous animal, and (2) the extremely low concentration of protein in the lymph from edematous animals. Although the difference between the protein concentrations of edema fluid and lymph from the same region is small, the conclusion is not yet justified that a similarly small difference exists between normal tissue fluid and normal lymph.

FROM THE AUTHORS' SUMMARY.

FATE OF BILIRUBIN IN THE SMALL INTESTINE. M. S. SACKLEY, C. G. JOHNSTON  
AND I. S. RAVDIN, *J. Exper. Med.* **60**:189, 1934.

Since there was no loss of bilirubin from the jejunal loop, and no loss of bilirubin when pigment was incubated with juice from the loop segment, or juice from the entire small intestine, it may be concluded that intestinal juice per se has no effect in converting bilirubin to urobilin in a two hour period, and that in the jejunal loop there was no absorption of pigment or conversion to urobilin. The experiments showing loss of pigment in the entire intestinal tract suggest that in some place other than the jejunal portion of the intestine the combined activity of the intestinal contents and intestinal cells does affect the bilirubin in the intestine. Whether the loss of bile pigment under such circumstances is due entirely to conversion, to conversion and absorption or to absorption of bilirubin as such remains to be answered by subsequent investigations.

FROM THE AUTHORS' SUMMARY AND CONCLUSIONS.

**EXPERIMENTAL HETEROTOPIC FORMATION OF DENTIN AND ENAMEL.** C. B. HUGGINS, H. R. McCARROLL AND A. A. DAHLBERG, *J. Exper. Med.* **60**:199, 1934.

The formation of dentin and enamel in the abdominal wall of the young pup was achieved by transplantation of the soft tissues of the developing tooth germ. An interesting finding was the cytomorphosis of the epithelium of the enamel organ. When this was transplanted so that the ameloblasts were in contact with the odontoblasts the cylindric character of the epithelial cells was preserved and enamel was produced; otherwise the cylindric shape of these cells was lost and a stratified epithelium resulted, resembling the gingival epithelium and certain tumors (the adamantinoma) of the jaw and related structures. This degenerated epithelium did not produce enamel and had an important characteristic of not forming cysts in a closed connective tissue space—forming, instead, islands and cords of cells with epithelial pearl formation. Thus the influence of mesodermic connective tissue derivatives on the form and function of epithelium is presented. The odontoblasts were found capable of survival as such and readily formed new dentin in transplantation; the stellate cells of the pulp were inert from the standpoint of inducing calcification.

FROM THE AUTHORS' SUMMARY.

**THE EFFECT OF THE GAMMA RAY OF RADIUM ON WOUND HEALING.** IRA T. NATHANSON, *Surg., Gynec. & Obst.* **59**:62, 1934.

Under standard conditions of distance and filtration but with varying dosages, the gamma ray of radium was used in the treatment of surgically produced wounds in dogs. Small and moderate doses produced an acceleration of healing in wounds exposed immediately. Greater amounts gave opposite results. In wounds 24 hours old, healing was always retarded, the degree varying directly with the dose. In those 48 hours old only the higher dosages employed slowed the healing process. Retardation did not interfere with the formation of a smooth scar.

FROM THE AUTHOR'S SUMMARY (W. C. HUNTER).

**INFLUENCE OF SPLENIC AUTOLYSIS ON THE LIVER.** N. FIESSINGER AND A. GAJDOS, *Ann. d'anat. path.* **10**:141, 1933.

The authors studied the changes following extensive crushing of the spleen in dogs and guinea-pigs. Blood and urine changes in general, such as those relating to urea, cholesterol, chlorides, etc., were of no significance, except that several days after the crushing of the spleen a marked bilirubinuria developed. This was traced to extensive degenerative and atrophic changes which developed in the hepatic parenchyma at that time. These changes were followed by focal scarring of the liver. But at the time when the traumatized splenic tissue underwent organization a second type of change in the liver occurred. This consisted of an extensive proliferation of the reticulum of the liver. This proliferation appeared suddenly and simultaneously with the organization of the crushed splenic tissue. The authors agree with Guy Albot that there are two types of reaction of the hepatic mesenchyma, namely, sclerosis and reticulosclerosis. Another interesting conclusion is that the mesenchyma throughout the body or in various organs may be stimulated by a single factor at the same time.

PERRY J. MELNICK.

**NORMAL AND PATHOLOGIC OSTEOGENESIS.** G. DUBREUIL, M. CHARBONNEL AND L. MASSE, *Ann. d'anat. path.* **10**:225 and 337, 1933.

The first paper is an extensive discussion of the classic and recent theories of osteogenesis. The essentials of the classic theory are as follows: In a vascular and connective tissue medium specialized cells appear, the osteoblasts, which elaborate osteoid tissue; the latter fixes calcium salts and thus becomes bone. Recent modifications of this theory by Heitz-Boyer and Scheikewitch and by Leriche and Policard have been widely accepted, especially by surgeons. Based on the fact that new bone can replace boiled bone pegs, bone fragments, etc., and

that transplanted periosteum does not form bone, these new theories relegate the osteoblasts to the background. Heitz-Boyer and Scheikewitch believe ossification to be a passive phenomenon which follows proper local concentration of calcium and depends on a preexisting inflammatory process. When the proper conditions irritate the periosteum (fracture, etc.) a proliferation of connective tissue results (vegetative osteitis). When this connective tissue finds a proper concentration of calcium it is transformed into bone. The cellular elements, therefore, have no specific character. Leriche and Policard have taken much the same view, and have analyzed in detail the various steps in the process. There are first an edematous loosening and modification of the connective tissue medium and other changes in the mesenchyma, which then becomes transformed into an osteoid tissue, the fibroblasts thus becoming osteoblasts without their having exerted any kind of specific influence. The preexisting local concentration of calcium is an important element in determining the steps in this process. The elaborate work of Leriche and Policard has gained wide acceptance.

Dubreuil, Charbonnel and Masse analyze each step in the theories in detail, and bring out various types of evidence to refute them. From the domains of embryology, histology, cytology and pathology they present a number of facts to prove that the osteoblasts are a specific type of cell, and that they elaborate within their cytoplasm a substance which is secreted and deposited as osteoid tissue, which then becomes calcified. They contend, therefore, that the essentials of the old classic theory of osteogenesis still hold.

In the second paper they report an extensive series of experiments which support the conclusion that the rôle of the osteoblasts is a specific one. The experimental work was done on dogs; various bones were incised, holes were bored, the periosteum was lifted or removed, with or without boring holes, pieces of bone were lifted or removed, and so on. Also, various bones were exposed to roentgen rays. Microscopic studies of the experimental material were made. In general, clearcut evidence of the specific activity of the osteoblasts could be seen. The authors therefore conclude that the classic theory of osteogenesis is still tenable. They credit Leriche and Policard and others with having stimulated much beneficial research and with having favorably influenced bone surgery, but they do not agree with their views as to the passive or inanimate nature of osteogenesis.

PERRY J. MELNICK.

#### EXPERIMENTAL NEPHRITIS. H. VASSILIADIS, Ann. d'anat. path. 10:703, 1933.

By injecting uranium nitrate, corrosive mercuric dichloride, cantharides or bismuth intravenously into rabbits Vassiliadis succeeded in producing renal lesions. These were acute, subacute or chronic, depending on the dose. He claims to have demonstrated glomerular lesions in the rabbits with ascites or anasarca, but no significant glomerular changes in those without water retention. In rabbits with slight water retention only a certain number of injured glomeruli were found. In most of the rabbits with toxic nephritis this condition was either not exudative or associated with only a very little exudate.

PERRY J. MELNICK.

#### EXPERIMENTAL STUDY OF CEREBRAL HEMORRHAGE. H. T. DEELMAN, Ann. d'anat. path. 10:977, 1933.

Two series of experiments were made to shed light on the question: Is cerebral hemorrhage the result of rupture of a blood vessel or due to hemorrhage by diapedesis following angiospasm? In one experiment a small amount of blood (0.5 cc.) was injected under pressure into the brains of several rabbits, thus imitating exactly the conditions of hemorrhage by rupture. The result was a massive hemorrhage which had the same characteristics as the human, namely, necrosis of the brain tissue, spreading of the blood along the perivascular spaces, necrosis of the smaller vessels and punctiform or annular hemorrhages at the periphery of the large one. In a second experiment finely ground glass suspended

in cocoa butter was injected into the carotid artery in several rabbits. The glass particles partially occluded the lumens of the smallest blood vessels, imitating the partial occlusion occurring in angiospasm. The result was typical cerebral hemorrhage resembling the former in every way. Deelman concludes that there is no single etiology. Both factors may produce the same results, in some cases rupture; in other cases, angiospasm.

PERRY J. MELNICK.

**APOPLECTIC ATTACKS AND THEIR PATHOGENESIS.** P. SCHWARTZ, Ann. d'anat. path. **10**:995, 1933.

Schwartz presents a clear discussion of the pathogenesis of cerebral apoplexy. In the middle of the nineteenth century Virchow's studies on embolism and thrombosis led him to conclude that these were the factors involved. Charcot and Bouchard in 1868 concluded that rupture of a cerebral aneurysm was a cause of hemorrhage. In the following half century the idea gradually became established that rupture of a blood vessel was involved. Pick and Ellis in 1910 invoked the idea of pseudo-aneurysms. But in 1918 Rosenblath established clearly that in the vast majority of cases of cerebral hemorrhage no ruptured blood vessel can be demonstrated. The classic work of Gustave Ricker of Magdeburg furnished the explanation. His experimental research showed that capillaries and arterioles are controlled by nerve impulses. Under the proper conditions, spasm of the blood vessels leads to anemia of the part supplied, then to capillary dilatation of the adjacent segment (parastasis), followed by hemorrhage by diapedesis from the dilated capillary. Attacks of cerebral vascular spasm in hypertension, then, result in encephalomalacia (anemic or nonhemorrhagic apoplexy) and, if more severe, to secondary diapedetic hemorrhage. This process explains the majority of cases of both nonhemorrhagic and hemorrhagic cerebral apoplexy in hypertension. Thrombosis, embolism and ruptured aneurysm are also found, but are rare.

PERRY J. MELNICK.

**PATHOGENESIS AND PHYSIOPATHOLOGY OF CEREBRAL HEMORRHAGE.** J. LHERMITTE, Ann. d'anat. path. **10**:1010, 1933.

The paper is a general discussion of the theories of the pathogenesis of cerebral hemorrhage and a short résumé of the physiologic basis for the clinical symptoms. Virchow, Charcot, Bouchard, Löwenfeld, Pick and Ellis are responsible for the theory that cerebral hemorrhage is due to the rupture of a blood vessel, either at the site of an atheromatous plaque or following necrosis of the wall of a blood vessel or rupture of an aneurysm. This theory has been strongly contested by Rosenblath, Westphal and Baer, Schwartz and others. The theory that circulatory changes, namely, vascular spasm, may be the cause, based on the work of Ricker, has gained much acceptance (Schwartz, Cohn and others). A third theory, that such hemorrhages are due to degenerative changes of the brain tissue, necrobiosis, has been proposed by Rochoux and others. A fourth theory considers that a combination of circulatory changes with preexisting degenerative changes of the brain tissue is responsible. Lhermitte favors the latter theory.

PERRY J. MELNICK.

**DEVELOPMENT OF NEW BLOOD VESSELS IN GRANULATION TISSUE IN A CELLULOID CHAMBER.** E. MANZ, Frankfurt. Ztschr. f. Path. **45**:464, 1933.

A method is reported by which one can study living granulation tissue with the aid of a transparent celluloid chamber implanted in the rabbit's ear. By this method it was found that new capillaries arise only from preexisting capillaries. There is no evidence that they can be formed by cells other than capillary endothelial cells. The opinion is expressed that the stimulating factor for the new formation of capillaries is not the local hyperemia but, very probably, a growth-promoting substance, apparently produced by leukocytes.

W. SAPHIR.

THE MECHANISM OF SECRETION IN THE THYROID GLAND. HARALD OKKELS,  
Acta path. et microbiol. Scandinaev., supp. 16, 1933, p. 303.

Cytologic studies of the thyroid gland were made in guinea-pigs stimulated with extract of the anterior lobe of the pituitary gland. Thirty minutes after intraperitoneal injection of this extract the thyroid cells began to swell enormously. One hour after the injection the Golgi apparatus became visible; at the same time there were vacuolation and emptying of the colloid. The metabolism rose rapidly during the first thirty to sixty minutes; after a slight decline it remained nearly constant at a rather elevated level. The degree of metabolic activity corresponded to the degree of enlargement of the Golgi apparatus. The mitochondria in the thyroid cells seem to play an important rôle during the formation of colloid. Iodine stimulates this feature of thyroid secretion but does not influence the Golgi apparatus. The mechanism of secretion in the thyroid gland is twofold; it comprises the formation and the absorption of colloid. The unique position of the gland from a histophysiologic point of view is due to its faculty of storing a provisional secretion outside the cells in larger quantities and for a longer time than any other gland. The mitochondria are considered responsible for the formation of the secretion, whereas the Golgi apparatus is involved in its ultimate discharge.

JACOB KLEIN.

A CASE OF PANMYELOSIS. G. de OLIVEIRA, Virchows Arch. f. path. Anat. 292: 203, 1934.

Oliveira gives the name "panmyelosis" to a condition characterized clinically and anatomically by an increase in erythrocytes, leukocytes and megakaryocytes. The clinical picture was that of erythremia or polycythemia vera; anatomically myelosis was the striking feature. A woman, aged 39 years, had had clinical symptoms for fourteen years. The erythrocyte count was 5,500,000; normoblasts were present in the peripheral blood. The leukocyte count varied at different periods from 10,000 to 110,000. The blood at the height of the illness contained 15 per cent myelocytes, 12 per cent promyelocytes and 6 per cent myeloblasts. In spite of a platelet count of 360,000 there was marked tendency to hemorrhage. The active formation in the bone marrow of erythrocytes and their precursors, myeloid cells and megakaryocytes, is interpreted as evidence of the origin of these three lines of cells from a primitive mesenchymal stem cell. Extramedullary formation of the three lines of cells in the lymph nodes and spleen was noted.

O. T. SCHULTZ.

HEMORRHAGIC THROMBOCYTHEMIA ASSOCIATED WITH ATROPHY OF THE SPLEEN.  
E. EPSTEIN AND A. GOEDEL, Virchows Arch. f. path. Anat. 292:233, 1934.

A man, aged 56 years, had been under observation for four years prior to his death. His first symptoms were bleeding from the gums and interstitial hemorrhages, especially of the thighs. At this time he had an erythrocyte count that varied from 5,000,000 to 7,250,000 and a leukocytosis of from 12,500 to 14,600 with a monocytosis of from 12 to 20 per cent and an eosinophilia of from 6 to 14 per cent. The platelets varied in number between 1,800,000 and 2,200,000; in stained preparations they appeared abnormal. In these preparations there were seen also Howell-Jolly bodies and nucleated erythrocytes. The condition at this time was believed to be polycythemia vera. In the course of the disease the erythrocyte count decreased, reaching 2,730,000 before death, with 1,700 nucleated erythrocytes. The terminal leukocyte count was 12,000. The platelet count remained well over a million throughout the course of the disease. The most striking finding at necropsy was a markedly atrophied spleen; it measured 4 by 2.5 by 1.5 cm. and weighed 7 Gm. Microscopically it consisted chiefly of sclerotic, very thick-walled arteries; between these was a small amount of collagenous tissue. The bone marrow contained many megakaryocytes. Hirschfeld described hematologic obsér-

vations similar to these following splenectomy. The atrophy of the spleen is ascribed to arteriosclerosis of this organ, the process being likened to that which takes place in the arteriosclerotic contracted kidney.

O. T. SCHULTZ.

EXPERIMENTAL STUDIES OF TISSUE LYMPH FLOW AND RESORPTION. H. LOESCHKE, Virchows Arch. f. path. Anat. 292:281, 1934.

The path of tissue lymph flow was studied by means of injections of colloidal solutions directly into various tissues and organs and into serous cavities, joints and the subdural space. Trypan blue was found most satisfactory for this purpose. Animals of a variety of species were used. The solution traveled along the spaces of the loose connective tissue. It flowed also along the fibers of dense connective tissue, the solution having a strong affinity for collagenous and elastic tissue. There was active flow through the fibrous capsules of the abdominal and thoracic organs. In the connective tissues the flow was reversible, and the color disappeared after a time, in marked contrast to the prolonged retention of the dye selectively stored in granular form by the reticulo-endothelial system. Parenchymatous cells sometimes stained diffusely; these were believed to be injured cells. Resorption occurred chiefly by way of pericapillary lymphatic spaces and was most active in tissues with a rich capillary network. Adipose tissue, wherever situated, was looked on as a most active and almost specific organ of resorption. Resorption was also active in voluntary muscle. During periods of capillary distention the fluid flow was from the blood stream into the pericapillary lymphatic; during contraction of the capillary the flow was in the opposite direction. The experiments furnished evidence that material injected into the peritoneal cavity is excreted by the liver, kidney and gastro-intestinal tract. The path of excretion was directly through the organ by way of the tissue spaces. The bearing of the experiments on the deposition of amyloid and hyalin, the spread of bacteria and the metastasis of tumor cells is briefly discussed. In chronic hydrops of serous or joint cavities, active resorption by the pericapillary lymphatics of adipose tissue leads to hypertrophy of this tissue.

O. T. SCHULTZ.

### Pathologic Anatomy

THE MYOCARDIAL ASCHOFF BODY. L. GROSS and J. C. EHRLICH, Am. J. Path. 10:467 and 489, 1934.

The clinical histories and anatomic material of seventy cases of uncomplicated rheumatic fever with Aschoff bodies in the myocardium were investigated. A classification of Aschoff bodies is suggested, based on the appearance and distribution of the collagen, argentophil fibers, cell cytoplasm and nuclei. This classification includes seven types of Aschoff bodies, which apparently bear some relation to the life cycles of the lesions. Each type is described and is considered to possess sufficient characteristic features to identify it as an Aschoff body specific for rheumatic fever.

It appears that these specific lesions pass through three stages in development. The earliest phases, represented by small cell coronal and reticular Aschoff bodies, have been found to occur up to the fourth week after the onset of the illness. The middle phases, represented by large cell coronal, syncytial coronal, mosaic and large irregular cell polarized Aschoff bodies, have been found to occur between the fourth and thirteenth weeks after the onset of the illness. The late phases are represented by polarized Aschoff bodies which occur from the ninth to the sixteenth week after the onset of the illness, and subsequently by fibrillar Aschoff bodies which occur after the thirteenth week of the illness. The earliest types of specific lesions are apparently influenced in their response by the reactivity of the tissue, depending on whether there has or has not been a previous attack of rheumatic fever, and also by the state of the collagen present in the interstices between the myocardial bundles. As a consequence, the evolution of the lesion may follow one or two main courses, determined by the initial lesion. The latter may

occur in the form of the reticular or the small cell coronal Aschoff body. The final phases of the life cycle of the Aschoff body are common to both main courses. With division of the material into four groups representing different clinical courses, there appears to be some change both in the incidence of the types of Aschoff bodies in the myocardium and in their localization. The observations reported here, however, can by no means be considered as furnishing sufficient statistical evidence on which to base final conclusions on this point. That the tempo of the life cycle may be considerably faster or slower than what has been described in this report seems very probable. Some of the stages in the model of the life cycle presented by us may be absent in some cases, abbreviated in others—or, indeed, may appear in an order the reverse of that which we have suggested. These facts can be determined with greater accuracy only after a much more extensive series of cases has been examined and, in the last analysis, must await confirmation by the hitherto unsuccessful transmission of this disease to animals. It is hoped, however, that further studies will be made along these lines in order that some of these interesting relations may be placed on a firmer footing.

FROM THE AUTHORS' SUMMARIES.

THE DISAPPEARANCE OF GLOMERULI IN CHRONIC KIDNEY DISEASE. A. R. MORITZ and J. M. HAYMAN JR., Am. J. Path. **10**:505, 1934.

The number of possibly patent glomeruli and glomerular scars has been estimated by a combination of injection and histologic methods. The average number of glomeruli in fourteen normal human kidneys was  $1,282,800 \pm 32,700$ . In chronic renal disease not only the number of patent glomeruli but the total number of recognizable glomerular structures was reduced. This was most marked in chronic glomerular nephritis. The number of possibly patent glomeruli frequently falls below 500,000 and may fall below 200,000. The total number of recognizable glomerular structures, including scars, was frequently below 600,000 and in some instances below 300,000. Since large numbers of glomeruli may disappear during the course of chronic renal disease, it is suggested that the final histologic pattern may not give as much information concerning the pathogenesis or severity of the disease as is commonly thought.

FROM THE AUTHORS' SUMMARY AND CONCLUSIONS.

THE STRUCTURAL CHANGES IN THE DIGESTIVE TRACT IN UREMIA. R. H. JAFFÉ and D. R. LAING, Arch. Int. Med. **53**:851, 1934.

The diphtheritic ulcerative processes which were encountered in 19.8 per cent of 136 cases studied could be traced to localized circulatory disturbances. The earliest changes consist of capillary hyperemia of the mucosa, increased production of mucus and dilatation of the small veins of the submucosa. The increased permeability which is associated with the extreme widening of the small blood vessels leads first to edema and later to hemorrhages. Bacteria from the intestinal content settle in the devitalized hemorrhagic areas of the mucosa and cause fibrinous exudation and necrosis. The necrotic parts are sequestered, and ulcers are formed. Attention is called to the occurrence of a pseudomembranous ulcerative colitis in uremia, the pathogenesis of which is identical with that of the intestinal lesions.

FROM THE AUTHORS' SUMMARY.

PNEUMOCOCCAL LIPOID NEPHROSIS AND THE RELATION BETWEEN NEPHROSIS AND NEPHRITIS. S. S. BLACKMAN JR., Bull. Johns Hopkins Hosp. **55**:1, 1934.

Evidence is collected from classic examples of lipoid nephrosis described in the literature and from the study of ten cases in children that nephrosis is a form of diffuse nephritis in which microscopic hematuria, secondary anemia of hemolytic origin and slight elevation of the blood pressure may all occur at times as part of the disease. Acute cases occur without insidious onset. In these both the

neutral fats and the doubly refractile cholesterol esters in the kidneys may be very meager. The nonprotein nitrogen of the blood is not constantly or progressively elevated.

The lesions in the kidneys consist chiefly of certain diffuse changes in the epithelium of the glomeruli and tubules, identical in kind with those which may occur in any nephritis. The disease may persist for a long time, at least in children, without the development of other lesions in the majority of the glomeruli. Scarred glomeruli may be found in small numbers. There is no evidence that these focal scars are secondary to changes in the tubules.

No changes in the glomerular capillaries can be recognized to account for the albumin in the urine. The most important histologic distinction between nephrosis and nephritis depends on the absence of coagula within the glomerular capsules in nephrosis; their presence in glomerulonephritis has not been fully determined, but seems obviously related to the excretion of fibrinogen in the urine in addition to albumin.

In the cases of nephrosis described, fat deposits were present in tissues other than the kidneys. In general, the amount of fat in the liver parallels that stained in the kidneys. The significance of the fat deposits in the tissues, including the doubly refractile cholesterol esters in the kidney, is entirely unknown. The presence of neutral fat and cholesterol esters is in no sense specific for lipoid nephrosis. In the kidneys the fat in the epithelium of the tubules of the cortex is found predominantly in segments of the convoluted tubules which lie in each cortical labyrinth. Although the epithelial cells of the tubules in the cortical rays show other alterations, varying from cloudy swelling to necrosis and regeneration, these cells contain scarcely any fat deposits. The edema of nephrosis cannot be explained by mechanical factors alone. There is good evidence that widespread capillary damage is one important factor in the pathogenesis of the edema.

There is no evidence that lipoid nephrosis is a metabolic disease. Data are presented which point to an etiologic relation between chronic pneumococcic infection and the pathogenesis of lipoid nephrosis in some cases. This is supported by the experimental reproduction of nephrosis in animals by means of pneumococcus toxin, which is to be described separately. **FROM THE AUTHOR'S SUMMARY.**

**TUMOR OF A SUBCUTANEOUS GLOMUS.** MICHAEL L. MASON and ARTHUR WEIL,  
Surg., Gynec. & Obst. **58:807, 1934.**

The tumor is rare, benign and nonrecurring and is characterized by painful crises radiating from a small bluish subungual or subcutaneous swelling on the extremities and by a peculiar angioma-like structure richly supplied with nerve fibers. Its resemblance to neurogenic tumor on the one hand and angioma on the other, together with the fact that special stains are required for the demonstration of the true nature of the growth, probably accounts for its not being recognized as an entity. The growth reported developed under the skin of the knee following an accident thirty-seven years previously; it was exquisitely painful, about 5 mm. in diameter, bluish red and connected with the underlying tissue by means of a blood vessel. It consisted of numerous blood vessels surrounded by epithelioid and spindle cells between which were isolated myelinated and unmyelinated nerve fibers. The authors regard the tumor as a hyperplastic glomus arising from a normal structure and for this reason urge that it be classified as a hamartoma or a hyperplasia rather than a blastoma.

W. C. HUNTER.

**STRICTURE OF THE RECTUM FROM INGUINAL LYMPHOGRANULOMA.** DAVID BLOOM,  
Surg., Gynec. & Obst. **58:827, 1934.**

The Frei test for inguinal lymphogranuloma was positive in three men and five women with rectal stricture. All were benefited by the usual treatment for this condition, so that it appeared fairly certain that the strictures were produced by the virus of inguinal lymphogranuloma. In four instances the lower part of

the rectum was involved while in the other four the whole rectum was affected. The fact that the genital lymphatics drain toward the rectum and the fact that this drainage differs in the two sexes explain the more frequent occurrence of rectal stricture and the lesser incidence of involvement of the inguinal nodes in women suffering from inguinal lymphogranuloma. The examination of rectal tissue from five patients revealed only simple or chronic productive inflammation. Opinion is divided as to whether stricture of the rectum associated with inguinal lymphogranuloma is due to lymphostasis with resultant fibrosis or is the direct effect of the virus. Bloom feels that most if not all of such conditions as esthiomene, anorectal syphiloma and so-called benign strictures of the rectum are identical and are due to the virus of inguinal lymphogranuloma.

W. C. HUNTER.

FENESTRAE AND POUCHES IN THE BROAD LIGAMENT AS A CAUSE OF STRANGULATED HERNIA. ARTHUR B. HUNT, Surg., Gynec. & Obst. 58:906, 1934.

This form of internal hernia is rare and probably is the least common of the intra-abdominal strangulated hernias. Only thirteen authentic cases of strangulation through defects of the broad ligament were found recorded in the literature, and only two cases in which such defects were present but unassociated with strangulation. This condition, however, probably is more common than the reported cases would indicate. In cases in which pouches were the associated defects, congenital anomalies may be strongly suspected. Distention and distortion of the broad ligament from pregnancy or pelvic tumors seem the most likely factors in the production of fenestrae in that structure. Older multiparae are almost exclusively affected, although nulliparous women are not immune. The Baldy-Webster operation may be looked on as an etiologic factor.

FROM THE AUTHOR'S SUMMARY (W. C. HUNTER).

AN EARLY HUMAN EMBRYO IN SITU. ROBERT TENNANT AND ELIZABETH M. RAMSEY, Surg., Gynec. & Obst. 58:968, 1934.

Macroscopically the site of the embryo appeared as a pale yellowish elevation 3 mm. in diameter and was surrounded by a halo of reddened endometrium. The primitive nature of the chorionic villi, the size of the blastocyst, the relative sizes of the amnion and yolk sac and the dimensions of the embryonic parts indicated that the embryo was of approximately the same age as the Peters embryo, namely, from 10 to 12 days.

W. C. HUNTER.

ENTEROGENOUS CYSTS. SAMUEL McLANAHAN AND HARVEY B. STONE, Surg., Gynec. & Obst. 58:1027, 1934.

Enterogenous or enteric cysts may occur along any portion of the gastrointestinal tract, but are most common in the ileocecal region and least common in the rectal area. They are composed of intestinal elements which may show great variations. Their origin is usually traced to the small diverticula of the intestine occurring in fetal life, and it is thought that the diverticula and cysts appearing later are different phases of the same process. Such an origin aids in explaining the location of the cysts with respect to the intestinal wall. Two cysts of this nature are reported, one in an adult of 48 years and one in an infant of 1 month. In each case the tumor lay in close association with the rectum and was successfully removed. Microscopically the cysts had the structure of rectum with some variations in the epithelial morphology.

FROM THE AUTHORS' SUMMARY (W. C. HUNTER).

NEPHROSIS OR NEPHRITIS? J. S. DUNN, J. Path. & Bact. 39:1, 1934.

The essential lesion in subacute hypotonic edematous nephritis is in the glomeruli and consists of abnormal permanent patency of their capillaries, which

may have resulted from previous inflammation. In accordance with the modern theory of renal activity, this lesion interferes with the kidney's excretion of water and salt by causing an imbalance of filtration and reabsorption, with the latter in excess. Albuminuria is a result of the same lesion and probably depends on the mechanical factor of dilatation rather than on abnormal permeability of the glomerular capillary walls. The high percentage of albuminuria is due to concentration in the tubules. The prominent tubular changes are secondary to the lesions in the tufts and have no important functional significance.

FROM THE AUTHOR'S CONCLUSIONS.

THE LATERAL GENICULATE BODIES AS VISUAL PATHWAYS. I. MACKENZIE, J. Path. & Bact. 39:113, 1934.

Research on cerebral localization during the past fifty years has proceeded mainly on the lines of an attempt to correlate the activities of motor and sensory organs with the functions of cortical areas, the structure and delimitation of which have been accurately defined. The present contribution is made at a time when attention is being directed to the structure and functional relationships of the intermediate seats of integration. Among these the lateral geniculate bodies have acquired a prominent place, and the evidence which has been adduced explains (1) their anatomic and physiologic disposition, (2) their relationship to the eyes and to the cerebral cortex, and (3) their participation in the construction of the special neural system which provides the anatomic substratum for visual sensation.

ON EPITHELIAL CYSTS OF THE RENAL PELVIS, URETER AND BLADDER. G. H. WILSON, J. Path. & Bact. 39:171, 1934.

Epithelial cysts in the mucous membrane of the renal pelvis, ureter and bladder are the result of chronic inflammation. Cystic pyelitis and cystic ureteritis are mainly the result of degenerative changes occurring in epithelial inclusions, but may also be derived from the closure of folds in the mucous membrane. Cystic cystitis usually affects the trigon and is probably chiefly caused by closure of pre-existing lacunae. Spontaneous healing may occur in some cases, and all traces of cysts and epithelial inclusions may disappear. The cysts or epithelial inclusions may be followed by tumor formation.

FROM THE AUTHOR'S CONCLUSIONS.

IS FATTY DEGENERATION OF THE HEART MUSCLE A PHANEROSIS? J. H. DIBLE, J. Path. & Bact. 39:197, 1934.

In all cases of fatty change in the heart which have been investigated in this study there has been an increase in fat in the effected portion of the muscle. The evidence indicates that this change is of the nature of a simple infiltration with depot fat. There is no evidence of a process of phanerosis.

FROM THE AUTHOR'S CONCLUSIONS.

THE PULMONARY FIBROSIS OF HAEMATITE MINERS. M. J. STEWART and J. S. FAULDS, J. Path. & Bact. 39:233, 1934.

A number of hematite miners in the West Cumberland mine field (Great Britain) have died with a grossly fibrotic lesion of the lungs in which silica and hematite dusts are present in large amount. Fifteen cases were investigated in the present study. All the patients died within the last three years. The lesion is a highly characteristic one, mainly on account of the hematite dust present, which causes the densely fibrosed areas to assume a bright brick-red color. The fibrosis is diffuse rather than nodular, often massive, and always most intense in the upper half of each lung. An associated or superimposed tuberculosis is the rule, being present in eleven of the fifteen cases. A characteristic clinical and roentgenographic picture is produced, and the diagnosis can usually be made with reasonable certainty a year or more before the fatal issue. In fourteen cases

the silica content of the lungs averaged 1.66 per cent of the dry weight. This may be compared with 1.72 per cent for a group of eight cases of ordinary silicosis and 1.78 per cent in a series of seven sandblasters examined by the same method. We are of the opinion that this lesion is a form of silicosis (siderosilicosis) resulting from the inhalation of silica-containing hematite dust generated by dry-drilling and shot-firing, and that serious trouble commenced only when the old "hammer and jumper" gave place to the dry mechanical drill. The use of the wet drill has diminished the risk to a certain extent only, as the speeding up of the drilling process has allowed blasting to take place at much shorter intervals, greatly increasing the dustiness of the atmosphere, as compared with the period prior to 1913. It is not possible at present to give even an approximate idea of the frequency of this condition. The number of persons at risk for varying periods of time during the past twenty years must run into some thousands, and it is clear from inquiry in the mine field that a certain proportion of them are suffering from this disease. The fifteen deaths from this cause recorded here occurred within the past three years, and it is unlikely that this includes all the deaths from siderosilicosis during that period.

FROM THE AUTHORS' CONCLUSIONS.

**ANATOMIC CHANGES IN THE DIAPHRAGM FOLLOWING PHRENICECTOMY.** W. S. STANBURY, Tubercl. 15:300, 1934. See also Am. Rev. Tuberc. 29:528, 1934.

In all but one of eleven cases the operation of choice was evulsion by the method of Felix. The duration of the paralysis varied from three weeks to six years. Atrophy of the diaphragm is evident as early as the third week after section of the phrenic nerve and is complete by the fourth month. After paralysis, one half of the diaphragm is elevated and eventrated into the thorax. With stretching it becomes a whitish membrane of parchment-like thinness. Histologically, the atrophy of the paralyzed half is complete and uniform. In one case only, a few normal muscle bundles were seen in one area, scattered among atrophic fibers. This probably represented an accessory nerve supply. There was marked distortion of the abdominal viscera in ten of the cases, in three of which a fatal gastroduodenal obstruction developed.

**HISTOLOGICAL CHANGES IN THE LIVER OF 66 CHINESE INFECTED WITH CLONORCHIS SINENSIS.** R. HOEPLI, Chinese M. J. 47:1125, 1933.

In sixty-six Chinese who for the greater part had met a more or less sudden death which was in no case apparently due to clonorchis infection, Clonorchis sinensis was accidentally found in the liver at autopsy. With the exception of one case the infection was always light or moderate. In the majority of the cases the larger bile ducts showed dilatation, thickening of the wall and formation of numerous glandular structures. Only two cases of hepatic cirrhosis were found, one of Laennec's type and one of portal cirrhosis corresponding in type to the cirrhosis parasitaria described by previous authors and probably due to clonorchis infection. Increase of periportal tissue in a varying degree was observed forty-nine times; infiltration with eosinophils, in thirty-seven cases; fatty changes of liver cells, frequently connected with atrophy of liver cells in the center of the lobules, was found twenty times. In five cases, the central veins were surrounded by new-formed fibrous tissue, and in four cases there occurred a thickening and hyalinization of the intima of small arteries. The results of the examination of the present material indicate that probably in many cases of moderate clonorchis infection in which the clinical symptoms are light or nonexistent already considerable histologic changes may be present in the liver.

FROM THE AUTHOR'S SUMMARY.

**DIAPHRAGMATIC HERNIA.** C. CONTAT, Ann. d'anat. path. 10:1, 1933.

A case of congenital true diaphragmatic hernia in a boy 18 months old is presented. The hernia was parasternal and bilateral. Only three similar cases have been

reported. A discussion of diaphragmatic hernia follows. Congenital false hernia forms over 86 per cent of the cases, is five times more frequent on the left side, and affects boys more frequently than girls. It results from a failure of fusion of the diaphragm during the third month of fetal life. Congenital true hernia is characterized by a sac composed of thinned-out diaphragm. Several theories regarding its formation include: diminished resistance of the diaphragm, asymmetrical development of the liver, and anomalous and insufficient blood supply to the diaphragm, with atrophy of the muscle fibers.

PERRY J. MELNICK.

**IDIOPATHIC HYPERTROPHY OF THE HEART.** J. C. POMPE, Ann. d'anat. path. 10:23, 1933.

A case of idiopathic hypertrophy of the heart in a girl 7 months old is described. The heart weighed 190 Gm. (normal 36 Gm.). Histologic examination revealed marked infiltration of the cardiac muscle fibers by glycogen. In addition, almost every other organ in the body was also infiltrated by glycogen. The literature on idiopathic hypertrophy of the heart is reviewed, and to the various theories of etiology another is added, namely, that the condition springs from a disturbance of glycogen metabolism. Seven cases recorded in the literature were similar histologically, but glycogen was demonstrated in only a few.

PERRY J. MELNICK.

**SUBCUTANEOUS PERIARTERITIS NODOSA.** K. LINDBERG, Arb. a. d. path. Inst. d. Univ. Helsingfors 7:159, 1933.

Two cases of periarteritis nodosa are reported in detail, and twenty-one cases recorded in the literature are reviewed. In some instances the vascular nodules are subcutaneous. In these the diagnosis may be aided by biopsy. The condition may occur in association with influenza, angina, erysipelas, suppurating wounds, diphtheria, articular rheumatism, gonorrhea and syphilis. It is considered by many to be a nonspecific hyperergic reaction to infection. The prognosis is serious because of possible involvement of the viscera. However, in one third of the reported cases the disease ran a benign course.

JACOB KLEIN.

**ATYPICAL LYMPHOGRANULOMATOSIS.** H. PFENNINGWERTH, Frankfurt. Ztschr. f. Path. 44:85, 1932.

Pfenningwerth believes that lymphogranulomatosis (Hodgkin's disease) may be atypical in regard to location, course and microscopic picture. It may involve the skin, spinal column, ribs, bone marrow, brain, dura, eye, parotid gland, nasopharyngeal space, thyroid, thymus, thoracic duct, trachea, lungs, pericardium, pancreas, liver, spleen (a completely isolated form), suprarenal glands, ovaries, uterus, placenta, testes, epididymis and prostate. It may take the course of cryptogenic sepsis, acute degeneration of the myocardium, infectious cholangitis, generalized tuberculous lymphadenopathy or lymphatic leukemia. Atypical lymphogranulomatosis may be characterized morphologically by a marked preponderance of lymphocytes, plasma cells, eosinophilic cells, epithelioid cells or giant cells. But it also may show only a small number of cells of any one of these types. The giant cells may resemble megakaryocytes or cells with phagocytic properties. As a result of irradiation with roentgen rays the epithelioid and giant cells may shrink and connective tissue fibers increase. Granulomas that consist of cells of the types seen in Gaucher's disease or that resemble diffuse reticulo-endotheliosis should not be considered typical lymphogranulomatosis. The diagnosis of atypical lymphogranulomatosis should never be made on the ground that any other somewhat similar disease can be ruled out. In other words, a diagnosis of atypical lymphogranulomatosis *per exclusionem* should not be made.

O. SAPHIR.

## Pathologic Chemistry and Physics

ACID-BASE BALANCE OF GASTRIC JUICE, BLOOD AND URINE BEFORE AND AT INTERVALS AFTER STIMULATION OF THE GASTRIC JUICE BY HISTAMINE.  
L. MARTIN, Bull. Johns Hopkins Hosp. 55:57, 1934.

A number of persons were studied to observe the electrolyte changes in the gastric juice, blood and urine during the period of gastric secretion. Histamine was used as a stimulant, and the gastric juice was continuously extracted during the period of observation. The subjects are described under two groups: (1) those who were able to secrete free hydrochloric acid into the gastric juice and (2) those who were not. The amount of salts lost from the body in group 1 was about four times that lost in group 2. In the blood of the first group the typical changes were a decrease of chloride and phosphate and an increase of carbon dioxide content and serum protein. There was a slight rise of total serum base. This represents a state of relative alkalosis. The urine became more alkaline in the majority of cases. Among the anions, chloride and phosphate fell while carbon dioxide increased. Of the cations, base, hydrogen ion concentration and ammonia nitrogen fell. In the group with achlorhydria the variations in the majority of the cases were similar in kind but different in degree from those in group 1. The difference in degree consisted of a smaller loss of electrolyte in the gastric juice and correspondingly smaller variations in the blood and serum. In the initial specimen certain distinctive differences between the groups were noted. In group 2 the carbon dioxide capacity of the serum more frequently fell and the blood chloride more often rose, although the rises were small. In the urine the change of  $p_H$  was apt to be less marked, and in a larger percentage of cases the urine became more acid or remained unchanged.

FROM THE AUTHOR'S SUMMARY.

A PHYSICO-CHEMICAL STUDY OF THE SACHS-GEORGI REACTION. E. M. DUNLOP AND S. SUGDEN, J. Path. & Bact. 39:149, 1934.

With nonsyphilitic serum, precipitation occurs in a limited zone of low concentrations of serum and electrolyte. It does not occur beyond this zone. With syphilitic serum, precipitation occurs in the same zone as with nonsyphilitic serum and also in a zone of high concentrations of serum and electrolyte. These two zones are essentially discontinuous. The precipitate obtained with syphilitic serum in the zone of low concentrations is similar in composition (nitrogen content) to the precipitate obtained with nonsyphilitic serum in the same zone, but different in chemical composition from the precipitate obtained with syphilitic serum in the zone of high serum and high electrolyte concentrations. The difference in behavior of syphilitic and nonsyphilitic serum in the Sachs-Georgi test is dependent on a qualitative difference between the two types of serum.

FROM THE AUTHORS' SUMMARY.

THE CONVERSION OF THE GLYCOGEN OF THE VAGINA INTO LACTIC ACID. R. CRUIK-SHANK, J. Path. & Bact. 39:213, 1934.

A series of observations and experiments was made to find out how the glycogen in the vaginal epithelium of infants and adult women is converted into lactic acid. The results indicated that the production of this acid is due principally to bacterial fermentation of the glycogen. Further, in vitro experiments have shown that Döderlein's vaginal bacillus, a member of the lactobacillus family, and it alone of the organisms likely to be present in the vagina as saprophytes or pathogens, is capable of directly fermenting glycogen with the production of lactic acid. Other lactobacilli, such as *B. bifidus*, *B. acidophilus-odontolyticus*, ferment glycogen late, after from seven to ten days' incubation, a delay which may be due to lack of habituation to this carbohydrate. On the other hand, *B. bulgaricus*,

which is used therapeutically in the treatment of vaginal discharges, failed to ferment glycogen. These findings do not explain the moderate degree of acidity in the bacteria-free vagina of the new-born infant or the lactic acid in hematocolpos fluid. An attempt to demonstrate a nonbacterial enzyme in vaginal secretion failed, but in view of the glycogenase in fresh serum, the glycogen in the vaginal cells may in the absence of bacteria be converted by such an enzyme to dextrose, from which in turn lactic acid is produced by a glycolytic cellular enzyme.

## FROM THE AUTHOR'S SUMMARY.

EXPERIMENTAL ASPERGILLOSIS OF THE SPLEEN. A. NANTA AND M. SENDRAIL, Ann. d'anat. path. **10**:677, 1933.

An experimental study was undertaken to determine if the scleropigmentary Gamma-Gandy nodules in the spleen in so-called mycotic splenomegaly are really of mycotic origin. The authors injected several varieties of Aspergillus intravenously and also locally into the spleen in dogs and rabbits. They were able to demonstrate that a number of species of Aspergillus heretofore considered innocuous are pathogenic. They were able to reproduce in the animals all the clinical features of splenic anemia, including the hemorrhagic, ascitic and icteric forms. Anatomically the lesions of this condition were also reproduced, including the splenomegaly, siderofibrotic nodules in the spleen, etc. Histologically the nodules were characteristic, with an outer hemorrhagic zone, a middle fibrotic zone containing giant cells and iron-filled macrophages, and an inner zone composed of long basophilic and hyaline mycelial threads. The lesions are probably formed by enzymes and local acidity produced by the fungus.

PERRY J. MELNICK.

QUANTITATIVE SPECTROSCOPIC ESTIMATION OF MANGANESE IN TISSUES. W. GERLACH AND K. RUTHARDT, Virchows Arch. f. path. Anat. **292**:52, 1934.

To Gerlach's series of contributions describing the quantitative spectroscopic elementary analysis of tissues the authors add a method for manganese.

O. T. SCHULTZ.

PEROXIDASE REACTION IN AN OVARIAN CYSTOMA. W. LOELE, Virchows Arch. f. path. Anat. **292**:135, 1934.

The columnar epithelium of some of the cystic and glandular spaces of a multilocular cystoma of the ovary was stained violet by a solution of alpha-naphthol and hydrogen dioxide. The reaction, which is dependent on the presence of naphthol peroxidase, is explained by degeneration of the cells in the presence of persistent leukocytosis.

O. T. SCHULTZ.

DISTRIBUTION OF MINERAL SALTS IN INCINERATED SECTIONS OF NECROSES AND ABSCESSSES. W. KLOSTERMEYER, Virchows Arch. f. path. Anat. **292**:268, 1934.

The distribution of mineral salts in ashed sections or spodograms of tissues containing necroses and abscesses prepared by the Schultz-Brauns method was studied by the indicator method of Hackmann. The ash of necroses consisted chiefly of insoluble calcium salts, together with probably sodium phosphate. When all nuclear material had disappeared from necroses alkaline carbonates could no longer be detected. The ash of abscesses contained relatively much potassium carbonate, less sodium carbonate and no insoluble calcium salts. Leukocytic infiltration of necroses led to a decrease in calcium salts and an increase in potassium carbonate. The potassium is probably derived from the nuclei of the tissue.

O. T. SCHULTZ.

THE ALBUMIN-GLOBULIN QUOTIENT IN ALBUMINURIA. J. BING, Acta path. et microbiol. Scandinav. **11**:323, 1934.

In examinations of the albumin-globulin quotient A/G in blood and urine the calculations ought to be made with the help of the relative albumin fraction (r. A. %), i. e., the fraction of the total protein which is represented by the albumin, instead of A/G, as this method affords much more exact values. The relative albumin fraction in the urine is dependent on two factors: (1) the ratio between the protein fractions in the blood and (2) the renal factor (R) which is calculated from the relation between the relative albumin fractions in the urine and blood. Examinations before and after transfusion of blood to patients with albuminuria, in whom changes in the relative albumin fraction of the blood of up to 20 per cent were detected, showed that the renal factor was constant. If the relative albumin fraction is particularly low in patients with amyloidosis, this is due to corresponding relations in the blood and not, as was hitherto supposed, to the circumstance that the glomerules, on account of their amyloid degeneration, are particularly permeable for the globulins. In the work of Hiller, McIntosh and van Slyke the renal factor was found to be high in the case of nephrosis and low in the case of chronic glomerulonephritis. The renal factor is never found to be lower than 1. The observations recorded in this paper are supportive of the modern conception of the mechanism of albuminuria as being an excretion of plasma proteins by filtration through injured glomerules.

FROM THE AUTHOR'S SUMMARY.

### Microbiology and Parasitology

THE PATHOGENESIS OF CHRONIC ULCERATIVE PULMONARY TUBERCULOSIS. E. R. LONG, Puerto Rico J. Pub. Health & Trop. Med. **9**:365, 1934.

Chronic ulcerative pulmonary tuberculosis, the adult type of pulmonary tuberculosis, or phthisis, represents reinfection, of progressive course, in a person who already has a primary focus of tuberculosis. The latter focus is commonly in the lungs, with secondary involvement of the tracheobronchial lymph nodes, but may be elsewhere; it represents a childhood type of tuberculosis that has become inactive or healed. The onset of chronic ulcerative pulmonary tuberculosis may be insidious, without symptoms, or acute, with symptoms resembling those of the commonly recognized acute respiratory infections. Anatomically it occurs in the apices of the lungs or in the subapical region and spreads downward. The downward extension takes place by intrabronchial spread from regions of ulceration in the upper parts. The essential condition for the whole course of progression is softening of the caseous tubercle. The mechanism of this softening is not exactly understood, but the process seems related to tissue hypersensitivity. Vast numbers of tubercle bacilli are present in softening caseous tissue, many more than in the walls of old cavities, and therefore each intrabronchial discharge of a softened mass leads to further spread within the lung, as well as to the outside world. This fact makes the softening of the caseous tubercle the key problem in the pathogenesis of tuberculosis.

FROM THE AUTHOR'S SUMMARY.

THE SINGLE PYOGENIC LIVER ABSCESS. ROBERT E. ROTHEMBERG and WILLIAM LINDER, Surg., Gynec. & Obst. **59**:31, 1934.

Cultures of eleven of the twenty-four abscesses investigated were sterile. Five yielded staphylococci. Streptococci, *B. mucosus-capsulatus* and unidentified gram-positive bacilli each occurred twice, while the colon bacillus and the pyocyaneus bacillus were each isolated once. The pathogenesis strongly resembled that of renal carbuncle and osteomyelitis in that nine persons had antecedent focal infection, suggesting hematogenous dissemination, probably by way of the hepatic artery. In eighteen of the cases the abscess lay in the dome of the liver. Surgical drainage was followed by recovery in 58.3 per cent of the cases.

W. C. HUNTER.

THE TREATMENT OF SEPTICAEMIA IN RABBITS WITH LYMPH-GLAND FIXATION ABSCESSSES. A. C. ALPORT, Brit. J. Exper. Path. **15**:175, 1934.

Fixation abscesses, obtained by the subcutaneous injection of desiccated lymph gland, were used for the treatment of septicemia in rabbits; the object was to cause leukocytosis, thus increasing the bactericidal power of the blood. Nine normal rabbits were used. Four were controls and received intravenous injections of virulent streptococci only; all died. The other five rabbits received similar doses of streptococci intravenously, but were also given subcutaneous injections of lymph gland; all these animals recovered. **FROM THE AUTHOR'S SUMMARY.**

THE INFLUENCE OF TEMPERATURE ON THE SURVIVAL OF PURE SUSPENSIONS OF THE ELEMENTARY BODIES OF VACCINIA. C. R. AMIES, Brit. J. Exper. Path. **15**:180, 1934.

Pure suspensions of the elementary bodies of vaccinia in a simple broth medium retain their activity for considerable periods at room temperature, and for several weeks at 37 C.

**FROM THE AUTHOR'S CONCLUSIONS.**

EFFECT OF SPLENECTOMY IN INFECTION WITH TRYPANOSOMA CONGOENSE IN MICE. C. H. BROWNING, D. F. CAPELL and R. GULBRANSEN, J. Path. & Bact. **39**:65, 1934.

The discrepancies in the effects of splenectomy in various protozoal and spirochetal infections in different species of animals and, in the case of *S. recurrentis*, apparently in infections with different strains of the parasite in the same host, suggest either that there is no common basis of defense against infections which seem to be similar in their pathology or else that the function of the spleen as an organ of defense must vary considerably according to the species.

A LUMINOUS ORGANISM IN RELATION TO NUTRITION ON AGAR. J. CRUIKSHANK, J. Path. & Bact. **39**:141, 1934.

A study was made of the factors concerned in the growth of colonies of a luminous organism on agar plates. By using the light produced by the colonies, recorded photographically, as a measure of their metabolic activities, it was shown that diffusible food substances are rapidly consumed from the agar in the neighborhood of the colonies and subsequently from agar at a distance. The volume of uninoculated agar from which the organisms may ultimately draw nourishment is the important factor in determining the amount of their growth. The same factor appears to be of importance in the colony growth of other organisms.

**FROM THE AUTHOR'S SUMMARY.**

THE BLOOD IN RATS AND MICE AFTER SPLENECTOMY, WITH OBSERVATIONS ON BARTONELLA MURIS AND EPERYTHROZOON COCCOIDES. J. A. W. MCCLUSKIE and J. S. F. NIVEN, J. Path. & Bact. **39**:185, 1934.

In mice removal of the spleen is occasionally followed by anemia and the appearance of bartonellae in the red corpuscles. The bodies are much smaller than those observed in the rat. A more frequent occurrence is the appearance of peculiar ring-shaped structures in the blood—Schilling's Eperythrozoon coccoides—some of which are free and some attached to the red cells. Attempts to infect both normal and splenectomized rats with eperythrozoon-containing blood have been without success. Attempts to isolate Bartonella muris and Eperythrozoon coccoides by various cultural methods have been unsuccessful. The evidence afforded by the morphology and staining reactions of the bartonella bodies of rats, by their incidence and behavior after splenectomy and by their transmissibility, as well as by the lesions accompanying their appearance, points to their being distinct from any of the tissue constituents of the host and suggests strongly that they

are micro-organisms. In the case of *Eperythrozoon coccoides* of the mouse the evidence is less strong but points in the same direction. The question is left open as to where these organisms are situated in the host during the latent periods.

CULTIVATION OF THE GONOCOCCUS. J. W. MCLEOD and others, *J. Path. & Bact.* **39**:221, 1934.

Cultural demonstration of the gonococcus is superior to demonstration by examination of smears in chronic cases of gonorrhea in both sexes and in all cases in females, specially when material for examination is taken from the cervix. There is, however, a residue of cases in which the smear is positive and the culture negative which, with the methods at present available, is larger than can be explained by falsely positive results. A small percentage of falsely positive results is undoubtedly recorded if diagnosis is determined by microscopic examination of Gram-stained smear preparations only. The cultivation of many strains of the gonococcus is prompted by incubation in air containing carbon dioxide in the concentration of 8 per cent. The recognition of gonococcal colonies in a culture is greatly facilitated by the use of the direct oxydase reaction, and the employment of this reaction results in a marked economy of time.

FROM THE AUTHORS' CONCLUSIONS.

THE ACTION OF EXTRACTS OF STAPHYLOCOCCI GROWN ON CELLOPHANE AGAR.  
LUISE BIRCH-HIRSCHFELD, *Ztschr. f. Immunitätsforsch. u. exper. Therap.* **81**:260, 1933-1934.

The culture medium of Jacobsthal is made by coating a thick layer of agar with cellophane. The crystalloid substances of the medium pass through the cellophane; the colloidal products of bacterial metabolism remain on the surface. Some bacteria grow abundantly on such medium, but they become quickly autolyzed. Birch-Hirschfeld used this technic for the study of staphylococcal hemolysin and protease. Hemolysin could be separated from the protease by precipitation with certain concentrations of acetic acid; both were precipitated with ammonium sulphate. Their thermostability was marked. Heating above 60 C. destroyed them gradually, but even after one-half hour at 100 C. from one twentieth to one tenth of the original titer was retained. Addition of dextrose to the culture medium interfered markedly with the formation of hemolysin, but had only a slight effect on that of protease; on the other hand, addition of blood enhanced the titer of the hemolysin; the production of protease was adversely affected, but only when the addition of blood amounted to 20 per cent or more. A normally nonhemolytic strain of streptococci became hemolytic when grown on cellophane agar, and another strain acquired marked toxicity under similar circumstances.

I. DAVIDSOHN.

SEASONAL FLUCTUATIONS OF SPONTANEOUS INFECTIONS IN GUINEA-PIGS. T. KJÄR, *Ztschr. f. Immunitätsforsch. u. exper. Therap.* **81**:511, 1933-1934.

Of 3,860 guinea-pigs which were treated with diphtheria toxoid during two years, 6.5 per cent died from intercurrent infections. The largest number of deaths occurred in March and April, which coincided with the minimum antibody response to diphtheria antigen. The smallest number of deaths was observed in August and October, which coincided with the optimum antibody response.

I. DAVIDSOHN.

### Immunology

THE VALUE OF THE NEGATIVE INTRACUTANEOUS TUBERCULIN TEST (MANTOUX) IN ADULTS. M. R. LICHTENSTEIN, *Am. Rev. Tuberc.* **29**:190, 1934.

Complete insensitivity to tuberculoprotein makes it certain that the patient is nontuberculous, with few exceptions (moribund or highly toxic patients, for a

variable period after harboring a contagious disease or after a course of tuberculin therapy). Patients who react only to strong concentrations of tuberculo-protein, with the same exceptions, almost certainly have no active tuberculosis.

H. J. CORPER.

THE RÔLE OF MULTIPLE REACTIVE GROUPS IN ANTIGEN-ANTIBODY UNION AS ILLUSTRATED BY AN INSTANCE OF CROSS-PRECIPITATION. M. HEIDELBERGER AND F. E. KENDALL, *J. Exper. Med.* **59**:519, 1934.

Antisera to R-salt-azo-benzidine-azo-crystalline egg albumin give precipitates with crystalline egg albumin by virtue of their antidyne content. The quantitative course of the reactions with increasing amounts of antigen is very similar for the dye-antidyne and egg albumin-antiegg albumin systems, but differs markedly for the cross-reaction between egg albumin and antidyne serum. A possible explanation for the occurrence of this one-sided cross-reaction is given in terms of reactive groupings on the antigen and antibody. A qualitative expression of the course of the cross-reaction is given in terms of the laws of classic chemistry.

FROM THE AUTHORS' SUMMARY.

FAILURE TO NEUTRALIZE POLIOMYELITIC VIRUS BY THE SERUM OF MACACUS RHESUS. N. P. HUDSON, E. H. LENNETTE AND E. Q. KING, *J. Exper. Med.* **59**:543, 1934.

Twelve specimens of serum from nine adult male monkeys failed to neutralize the virus of poliomyelitis. Ten samples of serum obtained from three adult female monkeys at various phases of the menstrual cycle likewise proved incapable of neutralizing the virus. An eleventh serum, drawn from a fourth female thirty-two days post partum, gave irregular results. It neutralized once and failed to do so on second test. This is the only suggestion in our experiments that a physiologic factor may play a part in immunity to poliomyelitis. Fourteen serums from ten immature monkeys caused to menstruate by treatment with anterior pituitary extract were devoid of virucidal property. This treatment failed also to induce a systemic resistance to intracerebral injections of virus in the nine monkeys of the same group available for testing. We were unable to demonstrate in our monkeys a correlation between the virucidal capacity of the serum and maturity or physiologic variations as exemplified by menstruation.

FROM THE AUTHORS' SUMMARY AND CONCLUSIONS.

SERUM DIAGNOSIS IN LEPROSY. J. LAIGRET, *Arch. Inst. Pasteur de Tunis* **22**:509, 1933.

A ground, heated and concentrated extract from the bacilli of rat leprosy was injected intradermally into four lepers and into controls. Three of the four lepers reacted, whereas three of the four controls failed to react. Complement-fixation tests were all positive, but syphilitic serum and tuberculous serum, especially the former, reacted in the few tests made. For this antigen, acetone and methyl alcohol were used to extract the rat leprosy bacilli. The possible value of negative serologic tests and the suggestion of antigenic similarity in human and rat leprosy were pointed out.

M. S. MARSHALL.

A TEST FOR THE CONFIRMATION OF THE SEROLOGIC DIAGNOSIS OF SYPHILIS. ERNST WITEBSKY, *Ztschr. f. Immunitätsforsch. u. exper. Therap.* **80**:323, 1933.

By means of Landsteiner's technic Witebsky separated the syphilitic antibody from the precipitate produced in the syphilitic serum with the citochol or Kahn antigens. The fluid with the antibodies, which was practically free from proteins, reacted positively in complement-fixation and flocculation tests. Sometimes a

positive reaction was observed when the native serum reacted negatively. That is explained by the elimination of inhibitory substances during the process of separation of the antibodies from the precipitate. The result adds the final link to the chain of proofs first furnished by Sachs, Klopstock and Weil that the changes in the syphilitic serum are an antigen-antibody reaction and not merely a result of colloidal imbalance. The procedure makes it possible to differentiate positive from certain falsely positive reactions. However, it will not help to differentiate syphilis from conditions in which lipoid antibodies are developed (trypanosomiasis, acute malaria).

I. DAVIDSOHN.

THE GROUP-SPECIFIC B-RECEPTORS AND THEIR ANTIBODIES. KURT MARBERG,  
Ztschr. f. Immunitätsforsch. u. exper. Therap. **80**:340, 1933.

Marberg reinvestigated the B quality in the red blood cells of the rabbit and man. He confirmed the previous report of Friedenreich and With by means of different procedures. The B quality in human red cells has some factors in common with the B quality in the rabbit cells, but they are not identical, as evidenced by the fact that the human anti-B ( $\beta$ ) iso-agglutinin was in most instances absorbed by rabbit red cells quite as well as by human cells of type B, while the anti-B hetero-agglutinin of rabbit immune serums, produced by inoculation of the rabbits with human B cells, was absorbed only by human B cells but not by rabbit cells. The latter phenomenon is in full agreement with established serologic facts. Occasionally human anti-B iso-agglutinin was not fully bound by rabbit red cells. Human saliva of group B had a powerful binding effect on the agglutinating properties of the rabbit anti-B immune serum. The binding was not affected by boiling.

I. DAVIDSOHN.

ACTIVE SUBSTANCES AND FUNCTIONAL CHANGES IN ANAPHYLACTIC SHOCK.  
W. ZECHNALL, Ztschr. f. Immunitätsforsch. u. exper. Therap. **80**:357, 1933.

Zechnall found an increase of potassium in the blood of shocked, actively sensitized guinea-pigs (from 25.7 to 43 mg. as compared with 21 mg. per hundred cubic centimeters in normal animals). A like increase was found during shock in passively sensitized guinea-pigs. The elimination of intravenously injected potassium in actively sensitized guinea-pigs was delayed. Contrary to the report of Jelin, intravenous injection of glycogen did not produce anaphylactic shock in the normal and in the sensitized guinea-pigs.

I. DAVIDSOHN.

INFLUENCE OF SNAKE VENOM ON THE HAPTENS. WALTER SCHEPERS, Ztschr. f. Immunitätsforsch. u. exper. Therap. **80**:395, 1933.

When alcoholic extracts of different types were mixed with weak dilutions of cobra venom, they failed to show the usual flocculation with antiseraums. Cobra venom alone did not inhibit flocculation if it was added to the mixture of anti-serum and alcoholic extract without preliminary incubation with the extract. Schepers explains the action as being due to the formation of nonspecific inhibitory substances by combination of the venom with certain lipoids of the haptens.

I. DAVIDSOHN.

EXPERIMENTAL ANAPHYLAXIS IN GUINEA-PIGS WITH PLANT ANTIGEN. P. MANTEUFEL and O. WICHELHAUSEN, Ztschr. f. Immunitätsforsch. u. exper. Therap. **80**:460, 1933.

Watery extracts of beans (*Phaseolus vulgaris*) and of peas (*Pisum sativum*) were injected intravenously into twelve rabbits. All the animals produced good precipitating serums. The older reports in the literature concerning normal precipitins against legumes were not confirmed. The method of passive anaphylaxis was a less reliable means than the precipitin test for the study of the antigenic

relations between the different varieties of the legumes. The Arthus phenomenon was less sensitive and less specific for the study of the sensitivity of guinea-pigs than was the active anaphylactic shock. The conjunctiva and the nasal mucosa of sensitized guinea-pigs did not react to local applications of the antigens. The Prausnitz-Küstner phenomenon was elicited in guinea-pigs sensitized with legumes but not in those sensitized with pollens. The seroreactions, the estimation of precipitins, the complement-fixation reaction, the passive anaphylaxis and the test of Prausnitz-Küstner are not to be depended on as reliable tests for the determination of sensitivity in guinea-pigs. Manteufel and Wichelhausen maintain that by the same token there is no justification for the strict separation of human allergy from experimental anaphylaxis in animals because the former fails to show some or all of the aforementioned serologic reactions. Attempts at desensitization of sensitized guinea-pigs were not successful. Administration of different anticoagulants and artificial production of fever did not prevent the development or decrease the intensity of the anaphylactic shock in actively sensitized guinea-pigs.

I. DAVIDSOHN.

### Tumors

DIBENZANTHRAcENE 1:2:5:6 AS A CARCINOGENIC AGENT. M. G. SEELIG, Am. J. Cancer **20**:827, 1934.

Seelig states that his work, on the whole, has confirmed the facts developed by the English workers. He found dibenzanthracene to be a cleaner, simpler, more manageable compound than tar. It is less toxic, and this makes possible the development of tumors in a larger percentage of mice than one can hope to secure with tar. On the other hand, the time necessary for the development of the tumors is markedly longer than with tar. When time is an important factor it might be much more desirable to use tar. Another disadvantage is the cost. The cost of high fraction tar is practically negligible, whereas Seelig paid \$3.50 a gram for dibenzanthracene.

FROM THE AUTHOR'S CONCLUSIONS.

SOMATIC MUTATION IN THE ORIGIN OF CANCER. M. R. CURTIS, W. F. DUNNING AND F. D. BULLOCK, Am. J. Cancer **21**:86, 1934.

The number of independent cysticercus tumors per host was related directly to the number of parasitic cysts. In the hosts with 1, 2 and 3 probably independent tumors the most frequent numbers of cysts per host were, respectively, from 1 to 6, from 6 to 11 and from 16 to 21. That is, an increase in the number of cysts per host was accompanied by an increase in the number of multiple primary cysticercus tumors. Both the cysticercus cysts and the cysticercus tumors were distributed to the several lobes of the liver in proportions approximately equal to the relative weights of the lobes. This indicates a chance distribution of both the cysts and the tumors. Rats which were infested with the larvae of *Taenia* and also inoculated subcutaneously with cysticercus sarcoma from another host sometimes acquired both a transplanted and a primary cysticercus tumor, sometimes neither and sometimes either one without the other. Indirect evidence indicates that the interval from the time the tumor could be recognized until it proved fatal was on the average about twenty-four days. The larvae from malignant cysts were not significantly longer than those from benign cysts of the same age, indicating that the sizes of the enclosed larvae were not a factor in determining which cysts became malignant. Of 4,321 cysticercus tumors, 79.3 per cent were sarcomas of the large cell type and probably arose from the cells which formed the inner zone of the cyst wall; 20.6 per cent were sarcomas composed of smaller cells and possibly arose from the outer zone of the cyst wall; 6 were adenomas, and 1 was a carcinosarcoma, which must have arisen from the snared-off bile-duct and hepatic epithelium which was embedded in the wall of the cyst. That is, the types of cysticercus tumors observed represent the types of cells found in the

cyst wall, and each type is represented by a number consistent with the expectation that the change of a normal cell to a tumor cell results from the chance action of an irritant.

FROM THE AUTHORS' SUMMARY.

CANCER CELLS IN THE BLOOD STREAM. E. H. POOL and G. R. DUNLOP, Am. J. Cancer **21**:99, 1934.

A hitherto undescribed large cell was found in smears of blood from seventeen of forty persons with cancer. Apparently the same cell was found in the blood of one noncancerous patient. The significance and origin of the cell are not established.

CONTAGIOUS LYMPHOSARCOMA OF DOGS. W. A. DEMONBREUN and E. W. GOOD-PASTURE, Am. J. Cancer **21**:295, 1934.

The tumor-inducing agent is destroyed by drying, freezing, glycerination and mechanical means. No evidence was obtained that a virus or any other infectious agent separable from the cells is concerned in the etiology of the disease. It is concluded that contagious lymphosarcoma is a true neoplasm and is transferable by the inoculation of living tumor cells in ulcerated surfaces. The origin of the tumor cells is not definitely determined, but they are probably derived from the lymphocytic series. The presence of neutral fat droplets in the tumor cell is recorded as characteristic. Multiple tumors can be induced by intravenous injections of the tumor cells in suspension. Growth of the tumor is associated with a variable immunity to reinoculation, and metastases seem to be related to periods of low resistance or absence of resistance which may occur during stages of massive and active tumor growth. A substantial immunity may be broken down by injection of large quantities of tumor cells. Serum obtained from rabbits immunized with emulsions of the tumor tissue is capable of destroying the tumor cells *in vitro*, and prevents the appearance of a tumor following inoculation of the treated cells. Serum obtained from rabbits immunized with normal dog serum affects tumor cells *in vitro* only slightly, and fails to prevent their growth when injected subcutaneously into dogs. The action on tumor cells of the heterophilic antibodies contained in such antiserums is negligible. Further studies are required to demonstrate definitely the presence of specific antibodies in the serum of animals immunized with emulsions of this tumor.

FROM THE AUTHORS' SUMMARY.

PITUITARY HORMONE IN CANCER. F. BISCHOFF, L. C. MAXWELL and H. J. ULLMANN, Am. J. Cancer **21**:329, 1934.

Sublethal doses of radiation to the pituitary gland, which brought about a temporary cessation of body growth, significantly retarded the growth rate of rat sarcoma R10, rat carcinoma 256 and a mouse carcinoma (spontaneous mammary) if the tumor appeared at the period of maximum retardation of the body weight. In the case of rat carcinoma 256, these effects were abolished by a simultaneous dosage of pituitary extracts with standardized growth-promoting powers. A cessation of increase in body weight produced by various poisons, equivalent to that following irradiation of the pituitary gland, failed to retard tumor growth significantly. In older rats dosage with standardized growth-promoting preparations of the pituitary gland significantly accelerated the rate of growth of carcinoma 256. In younger animals with mouse sarcoma 170 and rat sarcoma 10 the effect was less significant. Dosage of the urine of pregnancy augmented the effect of irradiation of the pituitary gland on the rate of growth of mouse sarcoma 180. The effect on rat carcinoma 256 and rat sarcoma 10 was less significant. Attempts to abolish permanently the function of the anterior lobe of the rat or mouse through irradiation of the pituitary gland were unsuccessful.

FROM THE AUTHORS' SUMMARY.

**EFFECT OF THE ANTERIOR PITUITARY HORMONES ON THE GROWTH OF MOUSE SARCOMA.** O. F. KREHBIEL, C. D. HAAGENSEN and H. PLANTENGA, Am. J. Cancer **21**:346, 1934.

Out of sixty treated animals fifteen survived the period of observation. Of these fifteen, seven had tumors which were slightly smaller than the tumors in the controls, while the remaining eight had tumors which were as large as, or larger than, those in the controls. Since the condition of all the treated animals suffered as a result of the injections, it would be expected that their tumors would be rather smaller than those of the untreated controls. These experiments fail to show, however, any specific and marked inhibitive action of anterior pituitary hormone on tumor growth as claimed by Zondek and his collaborators. It should be noted that it has been impossible to demonstrate an inhibitive action of anterior pituitary hormone on tumor growth despite the use of doses of the hormone which are so enormous in terms of the amount of the hormone normally excreted in the adult human being that all calculations become relatively meaningless. Katzman and Doisy have calculated that the average daily prolan excretion of adult males is 8 mouse units and that of adult females 10 mouse units. Yet 200 mouse units a day did not inhibit tumor growth in mice. These facts should discourage any attempts to use anterior pituitary hormone in the treatment of human cancer.

## FROM THE AUTHORS' DISCUSSION.

**DOES CHICKEN BLOOD PRODUCE IMMUNITY TO RAT TUMORS?** C. D. HAAGENSEN, Am. J. Cancer **21**:376, 1934.

The intraperitoneal injection of heparinized chicken blood in young rats does not produce immunity to inoculation with Walker rat carcinoma 256.

## FROM THE AUTHOR'S CONCLUSIONS.

**VENEREAL SARCOMA OF THE DOG.** E. L. STUBBS and J. FURTH, Am. J. Path. **10**:275, 1934.

Two venereal sarcomas were successfully transmitted to healthy dogs, and one of them was transplanted in eleven successive generations. Inoculations were successful in 72 per cent of the dogs given subcutaneous injections of an emulsion of tumor cells. Tumor appeared at the site of inoculation within an average of thirty-eight days and with one exception began to regress after reaching a size of about 10 cm. in the longest diameter. In one dog the tumor spread by metastasis throughout the body. Intravenous inoculation produced generalized sarcomatosis in two of seven inoculated animals. Transmission was also successful on rubbing tumor material into the scarified surface of the glans penis. Attempts to transmit the disease through intact mucous membrane (the conjunctiva) were unsuccessful. The ability of the tumor material to transmit the disease was destroyed by the addition of 50 per cent glycerin, by desiccation, by freezing and thawing and by heating to 50 C. for one and one-half hours. Tumor material passed through siliceous filters likewise failed to produce tumors. These experiments indicate that venereal sarcoma, often designated "infectious sarcoma of dogs," is a neoplastic process and like other mammalian tumors can be transmitted only by viable tumor cells.

## FROM THE AUTHORS' SUMMARY AND CONCLUSIONS.

**THROMBOPENIC PURPURA IN CARCINOMA OF THE STOMACH WITH METASTASES.** J. S. LAWRENCE and E. B. MAHONEY, Am. J. Path. **10**:383, 1934.

It would seem as though the presence of large numbers of tumor cells in the marrow was the probable cause of the thrombopenia, although normal megakaryocytes were present. Increased destruction of platelets in the puerperal circulation cannot be excluded.

"HEAVY WATER" AND TUMOR GROWTH. W. H. WOGLOM AND L. A. WEBER, J. A. M. A. **102**:1289, 1934.

Deuterium, in the amounts that it was possible to administer as "heavy water," had no demonstrable effect on the growth of mouse sarcoma 180 or mouse carcinoma 63.

FROM THE AUTHORS' CONCLUSION.

CHANGES IN THE ESTERASE AND FAT CONTENT OF THE SERUM INDUCED BY CANCER AND CANCER-PRODUCING AGENTS. H. N. GREEN, Brit. J. Exper. Path. **15**:1, 1934.

During the growth of Jensen sarcoma of the rat the esterase content of the serum falls progressively, ultimately reaching a very low level. The esterase content of the liver, lungs and kidneys is also much diminished. The phosphatase content of the serum also falls, but the average fall is less than half that of the esterase. In rats resistant to inoculation of the Jensen sarcoma the esterase content of the serum tends to rise. The fatty acid content of the serum rises in many and possibly in all rats during the growth of the Jensen sarcoma. It may reach a maximum level approaching 2 per cent, and then falls during the terminal stages of the tumor's growth. There is an associated rise in the cholesterol, but to a much less degree. With tar epitheliomas of mice and localized carcinomas of man the serum esterase content ranges around the normal, with a tendency to rise slightly. Evidence was obtained that the application of tar or the inoculation of tar or of 1:2:5:6-dibenzanthracene produces a rise in the esterase content of the serum of rabbits in a proportion of the cases.

FROM THE AUTHOR'S SUMMARY.

### Medicolegal Pathology

BLOOD GROUPING IN FORENSIC MEDICINE. L. LATTEs, Ann. d. méd. lég. **14**: 245, 1934.

A review of the literature reveals that the heredity of the agglutinogens M and N has been studied in 1,039 families with 2,900 children. Furthermore, in tests on 3,751 mothers with 5,912 children there was not a single exception to the theory of Landsteiner and Levine. Hence, the medicolegal application of the agglutinogens M and N for the exclusion of paternity is justifiable at present.

On the other hand, because of the difficulties in the technic and irregularities in the heredity, the subgroups of group A and group AB are of value only as confirmatory evidence for disproving paternity.

A. S. WIENER.

PRECIPITIN REACTION OF TEETH. H. PLATHNER, Deutsche Ztschr. f. d. ges. gerichtl. Med. **23**:61, 1934.

Extracts from finely comminuted human and animal teeth give species-specific precipitin reactions. Intact human molars and wisdom teeth are exceptions; also enamel. Teeth long buried in the ground or exposed to the air may not give any reactions.

DEMONSTRATION OF GROUP-SPECIFIC SUBSTANCES IN BODY FLUIDS. G. STRASSMANN, Deutsche Ztschr. f. d. ges. gerichtl. Med. **23**:186, 1934.

Strassmann's studies corroborate the findings of other investigators with regard to the presence of group-specific substances in body fluids. He found iso-agglutinins present post mortem in transudates in the pericardial, pleural and peritoneal cavities, rarely in saliva and never in cerebrospinal fluid. Receptors, however, are present not only in transudates, but in saliva, semen, gastric secretions, urine, meconium, vaginal mucus, nasal secretion, sweat and stains of all these secretions. The receptors may be demonstrated by the ability of the stain to inhibit

specifically the iso-agglutinins in a group O serum. A control test with unstained material must be made to rule out the possibility of nonspecific inhibition. The receptor peculiar to group O blood may be directly demonstrated by inhibition tests with ox serum previously absorbed with human blood of group AB. The fact that in the dry state the receptors resist physical and chemical agents for periods of months or years increases the value of the tests. One must always bear in mind that not every person secretes the receptors.

A. S. WIENER.

**DEMONSTRATION OF CARBON MONOXIDE POISONING 144 DAYS AFTER DEATH.**  
P. HEILMANN, Deutsche Ztschr. f. d. ges. gerichtl. Med. **23**:215, 1934.

It is well known that carbon monoxide has been found several weeks and months after death in disinterred bodies. A case is described in which carbon monoxide was found 144 days after death. The body was that of a man, aged 55 years, who was found dead in the morning in a room which contained two coke ovens. In spite of considerable decomposition the cherry red color was marked in the subcutaneous fat, the muscles, the blood and the organs. Carbon monoxide was demonstrated chemically and physically. No other cause of death was found.

**STRUCTURAL CHANGES IN ACUTE AMMONIA POISONING.** G. I. VON FAZEKAS,  
Deutsche Ztschr. f. d. ges. gerichtl. Med. **23**:225, 1934.

Ammonia acts not only as a local corrosive but as a general poison which is rapidly absorbed. In addition to laking the red cells of the blood it injures the endothelial lining of the blood vessels severely, and fatty changes develop in the internal organs, especially the liver and the kidneys. Marked lipemia may result. In the central nervous system severe alteration develops, especially in the ganglion cells of the cerebral cortex, but also of the white matter.

**MAJOR PROBLEMS IN BLOOD GROUPING DURING THE PERIOD 1927-1933.** L. HIRSZFELD, Ergebni. d. Hyg., Bakt., Immunitätsforsch. u. exper. Therap. **15**: 54, 1934.

This review supplements Hirsfeld's previous review on the same subject published in the same journal in 1928. The topics presented are: the heredity of the four blood groups, the subgroups of group A and group AB and their heredity, panagglutination, development of the blood groups, group-specific substances in the organs and body fluids, the agglutinogens M and N and their heredity, medico-legal applications, and individual differences in animal blood. The principal value of this review lies in its comprehensive nature and extensive 50 page bibliography. The review suffers, however, from the lack of emphasis on technic, and because the selection of material is uncritical.

A. S. WIENER.

**UNEXPECTED AND SUDDEN DEATH IN CHILDHOOD.** S. A. SIWE, Upsala läkareförh. **39**:203, 1934.

This article is based on the study of 212 cases of unexpected or sudden death in the first fifteen years of life. Most of the cases occurred in the first months, and in the majority death was due to diseases of the respiratory organs. Such diseases may not give rise to local symptoms and are easily overlooked. Even at the necropsy the extent of the infectious lesion may be limited. In nurslings a simple coryza or a slight intestinal disturbance may lead to vomiting and aspiration, which rapidly and sometimes unnoticed end in death. In none of the cases studied did enlargement of the thymus appear to play any rôle in causing death. The possibility of respiratory aspiration does not seem to be recognized sufficiently. At the necropsy it is not enough to inspect the finer bronchi for gastric contents. Litmus paper must be used in order to be sure that aspiration of stomach contents is not overlooked.

## Technical

BACTERIAL CAPSULES AS DEMONSTRATED BY A SIMPLE METHOD. J. W. HOWIE and J. KIRKPATRICK, *J. Path. & Bact.* **39**:165, 1934.

A simple and reliable method for the demonstration of bacterial capsules consists in adding to a loopful of exudate or of culture suspended in broth on a microscope slide first a drop of dilute carbolfuchsin, followed by a drop of 5-10 per cent solution of eosin, and then making films. The bacterial bodies are positively stained, and the capsules are seen by "relief staining." Suspending capsulated organisms in water hinders or completely prevents the demonstration of the capsules but does not destroy them. Capsules and bacterial bodies can still be demonstrated in cultures of pneumococci treated by addition of bile salt.

FROM THE AUTHORS' SUMMARY.

METHODS OF STUDY OF PULMONARY SILICOSIS. E. BEHR, *Ann. d'anat. path.* **10**:849, 1933.

The chief histologic methods of demonstrating the minute quartz crystals in pulmonary silicosis are discussed. Watkins, Pitchford and Moir in 1916 recommended acid digestion of the tissues. Policard and Okkels used incineration, which has the advantage of retaining the relationship of the crystals to the tissues. Giese in 1931 recommended mounting mediums for the sections which have different indexes of refraction from quartz. Behr used Giese's methods in the study of a case and found them excellent. He was surprised at the large numbers of crystals which could be seen. Three photomicrographs illustrate the relationship of the crystals to the tissues.

PERRY J. MELNICK.

A QUALITATIVE TEST FOR BARBITURATES IN THE URINE. W. MOHRSCHULZ, *München. med. Wchnschr.* **81**:672, 1934.

Poisoning with barbiturates, accidental or intentional, has increased during the past few years so as to be the commonest of all medicinal poisonings. These compounds appear promptly in the urine, where their detection is important in establishing barbiturate poisoning. Mohrschulz describes a simple rapid qualitative test for demonstrating barbiturates in the urine: From 15 to 20 cc. of urine is vigorously boiled with 0.2 Gm. of charcoal, centrifugated hot, the aqueous layer poured off, and the centrifuge tube dried with filter paper. The charcoal is washed into a reagent glass with 3 or 4 cc. of absolute alcohol, extracted with 7 cc. of chloroform and the mixture warmed, shaken and filtered. About 2 cc. of the filtrate (usually turbid) is clarified with small additions of absolute alcohol. To this mixture are added 20 drops of a 1 per cent cobaltic nitrate solution in absolute alcohol and then, drop by drop, a 1 per cent solution of potassium hydroxide in absolute alcohol. A dark blue coloration indicates the presence of a barbiturate.

EDWIN F. HIRSCH.

THE USE OF THE CENTRIFUGE IN THE M. K. R. II (MEINICKE CLEARING REACTION) FOR THE DIAGNOSIS OF TUBERCULOSIS, SYPHILIS AND GONORRHEA. F. E. HAAG AND AGNES DANE, *Ztschr. f. Immunitätsforsch. u. exper. Therap.* **81**:101, 1934.

The latest modification of the Meinicke precipitation test for syphilis is based on the use of a new extract and on the employment of the centrifuge. The extract contains the ether-insoluble, but alcohol-soluble fraction of beef heart to which balsam of Tolu and Victoria blue are added. A 3.5 per cent solution of sodium chloride is used as a diluent. The mixture of serum and of diluted antigenic extract is shaken and then centrifugated at 2,000 revolutions for ten minutes. The supernatant fluid is discarded, the tubes turned upside down, and the result is estimated from the appearance of the blue sediment. The same extract was applied by

Meinicke for the diagnosis of malleus, infectious abortion and gonorrhea. For that purpose the specific bacterial antigen is added to the sodium chloride solution before it is mixed with the extract. In a study of 5,239 serums, the results of the new Meinicke test were compared with those of several complement-fixation and precipitation tests for syphilis. It was found more satisfactory than the older procedures of Meinicke, and the results compared very closely with those of Müller's ball test. Its rapidity and the ease with which the results can be read are in its favor. The reaction was tested on a series of patients with gonorrhea, and it was found slightly more sensitive than the complement-fixation test but only in cases without complications; otherwise the complement-fixation test was preferable. It could not be used for purposes of differentiation in patients who also had syphilis, because then it was always positive. The new procedure was also employed for the diagnosis of tuberculosis. The antigen of Witebsky, Klingenstein and Kuhn (a benzene extract of tubercle bacilli) proved most satisfactory of a number which were tried. The results were better than any so far reported, particularly with regard to cases of tuberculosis of bones and joints, in which the previous methods were usually very disappointing. No negative results were obtained in fifty-nine clinically established cases of pulmonary tuberculosis, and on the other hand there were no positive results in thirteen cases in which the disease was known to have been clinically cured. In negative control cases, only 3.6 per cent of the results were falsely positive. The disadvantage of the method is that all patients with syphilis react positively.

I. DAVIDSOHN.

THE KAHN TEST MODIFIED BY CENTRIFUGATION. CARL SCHLESMANN, Ztschr. f. Immunitätsforsch. u. exper. Therap. 81:467, 1934.

According to Schlesmann, there is no real need for three tubes as they are used in the standard Kahn test. He uses the middle tube only (with 0.025 cc. of the antigen), adds 0.15 cc. of serum, shakes the mixture vigorously, and places it for twelve minutes in the incubator at 37 C. (or for four minutes in the water bath at the same temperature). Then the test tube is again shaken and centrifugated for ten minutes at 2,200 revolutions. After the addition of 0.5 cc. of physiologic solution of sodium chloride, the test tube is again shaken, and the result is read. Schlesmann carried out 2,400 tests (along with the official Wassermann test, the complement-fixation test which employs the very sensitive pallida antigen, the Sachs-Georgi test and the third Meinicke precipitation test) and found that the reading was made easier and that the test was more economical than, and at least as sensitive as, the standard Kahn procedure.

I. DAVIDSOHN.

# Society Transactions

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## NEW YORK PATHOLOGICAL SOCIETY

*Regular Meeting, March 27, 1934*

WILLIAM C. VON GLAHN, *President, in the Chair*

### A CASE OF DIABETES INSIPIDUS WITH AUTOPSY. LAWRENCE SOPHIAN.

A white woman, 33 years old, began to suffer with nausea and vomiting in August 1933. This continued, and she lost strength and weight. Clinical examination showed nothing unfavorable except a low grade fever and leukocytosis for about a week, with subsequent improvement. Polyuria and polydipsia were present, and the specific gravity of the urine was persistently between 1.002 and 1.010. There was some albuminuria, but the urine contained very few formed elements. The great weakness suggested a diagnosis of Simmonds' disease. The fluid output was from 3,800 to 7,600 cc. The intake and output were reduced by the injection of a solution of pituitary, but the patient apparently felt worse under these conditions. Roentgen examination of the bones and thorax revealed nothing. The blood chemistry, blood pressure and eyegrounds were normal. The clinical course showed an increase in apathy and a terminal subnormal temperature. The patient died after two months in the hospital.

Postmortem examination of the body showed all organs normal except the bones, the pituitary gland, the heart and the kidneys. The thyroid and suprarenal glands were examined and appeared normal with the exception of a very small fetal adenoma in the thyroid. Small streaks of opacity found in the heart and kidneys proved, on section, to be zones of necrosis with numerous gram-positive cocci in them microscopically. This appeared to be an agonal phenomenon.

The pituitary gland measured 17 by 12 by 9 mm. Near the stalk posteriorly was observed a projecting clear nodule 2 mm. in diameter. Serial sections were made through the gland and through the neighboring base of the brain, including the third ventricle. The main cyst and several smaller cysts were found to be in the pars intermedia, and were lined by simple low cuboidal epithelium. The smaller cysts contained colloid. In the neighboring portions of the posterior lobe no necrosis or proliferation of glia could be observed. The posterior lobe showed invading columns of basophilic cells. The constitution of the anterior lobe appeared normal in all portions. No ischemic or necrotic lesions were found in a search of 140 sections.

The bones on section presented a peculiar combination of osteolysis and osteogenesis. The gross change which brought this to notice was a granular roughening of the outer surface of the skull, which had a reddened appearance. On section the diploe was absent, and the bone tissue appeared softer than normal, as were the ribs and vertebrae. The type of bone change does not appear to be specific of any disease. The bone trabeculae were broadened, and the marrow channels were filled with fibrous tissue containing numerous hyperemic vessels. A considerable accumulation of osteoblasts could be found in some of these spaces. There was also new bone formation in small peripheral islands fairly well demarcated from the older bone.

The several features of this case do not appear to form any of the hitherto described syndromes. Clinically the weakness and cachexia were pronounced, but did not have the appearance of progeria as characteristically seen in Simmonds' disease. The response to the medication with pituitary solution was moderately

satisfactory in causing concentration of the urine, but the symptoms seemed to be aggravated. Histologically the basophilic infiltration of the posterior lobe appears to be the same process described recently by Cushing as an indication of activation of the posterior lobe. This would appear to be contradictory to the deficiency hypothesis ordinarily held in diabetes insipidus. As far as the bone changes are concerned, they do not appear to be similar to those in Paget's disease or to those in any of the clinical or experimental cases of hyperparathyroidism.

#### DISCUSSION

IRVING GRAEF: I should like to ask if the parathyroid glands were examined.

LAWRENCE SOPHIAN: I made a search for them and could not find them. I took several small pieces of tissue which I hoped were parathyroid glands, but I could not identify them afterward.

#### ANEURYSMS OF THE SINUSES OF VALSALVA: REPORT OF A CASE. H. D. KESTEN.

A colored youth, aged 19, who stated that he had had a chancre one and one-half years previously, followed by a rash, was admitted to the Presbyterian Hospital with intermittent complete heart block associated with an enlargement of the heart and pulmonic and apical systolic murmurs. The Wassermann test was negative repeatedly. An acute febrile polyarthritides developed following a hemolytic streptococcal angina. Death occurred two years after the appearance of the chancre, with signs of cardiac failure.

The postmortem examination was unfortunately limited to the heart. This organ was enlarged symmetrically and contained two aneurysms. One, the larger, opened by a buttonhole mouth into the base of the right aortic sinus and extended downward and posteriorly for several cubic centimeters into the interventricular septum. The other, occluded by a recently formed thrombus, lay beneath the left sinus, communicating with it. The coronary arteries were not compromised. The aortic valve cusps and arch of the aorta were normal. In the myocardium about these aneurysms were extensive areas of caseation, necrosis, numerous obliterated arterioles and collections of lymphoid and plasma cells. No spirochetes were found. The myocardium also contained characteristic Aschoff bodies. In addition, a penile scar was present. The aneurysms apparently developed as outpouchings into tissues which were necrotic and softened, probably as the result of the inflammatory obliterating endarteritis—syphilitic or rheumatic—of numerous septal arterioles.

#### FATAL ESTIVO-AUTUMNAL MALARIA IN DRUG ADDICTS IN NEW YORK CITY. MILTON HELPERN.

Twenty-one fatal cases of estivo-autumnal malaria of the cerebral form and one case of quartan malaria were examined post mortem by members of the staff of the Chief Medical Examiner. The fatalities occurred in a total group of forty-nine cases of malaria of which thirty-nine were of the estivo-autumnal type, nine were quartan and one was tertian, during a period from Sept. 25, 1933, to Feb. 28, 1934. In every instance the patient was a drug addict of the type employing the intravenous route for the injection of the drug. The disease was transmitted directly from addict to addict by the common use of unsterilized syringes.

Twenty of the persons who died of estivo-autumnal malaria died in coma and one of lobar pneumonia which developed several days after the temperature had dropped to normal and the parasites had disappeared from the blood. One who had quartan malaria displayed maniacal symptoms and died of bronchopneumonia.

The pathologic changes were fairly constant. In all the cases of fatal estivo-autumnal malaria in which coma developed the plasmodia were localized in great numbers in the capillaries of the brain. Parasites were not found in the brain of the patient who died of lobar pneumonia or in the brain of the one who had fatal quartan malaria. No excessive accumulation of malarial parasites was found in the capillaries of other organs.

The cerebral findings, the soft, swollen and only moderately enlarged spleens and the lack of pigmentation in the bone marrow indicated an acute course of the disease. The localization of the parasites in the brain in all the fatal cases suggested the possibility of a single strain of *Plasmodium falciparum-quotidianum* with an affinity for the cerebral capillaries. This was consistent with the epidemiologic findings, which indicated that the disease, after having been introduced by a carrier, was directly transmitted from addict to addict.

A complete report of the clinical, pathologic and epidemiologic observations in this series of cases is in preparation.

#### DILATATION OF THE PULMONARY ARTERY. B. S. OPPENHEIMER.

*Idiopathic Dilatation of the Pulmonary Artery.*—A study was made of seven cases which presented very similar clinical and roentgen features, which indicated a huge dilatation of the whole pulmonary arterial tree. Two of the patients came to necropsy, but no cause for the enormous dilatation of the pulmonary artery and its branches could be found. The finding of a similar dilatation in a 5 month old infant by Zuber suggests that such a dilatation may be primary and congenital, and the atherosclerosis, when it occurs, secondary. The diagnosis may be suspected clinically from the cyanosis, dyspnea, cough, secondary polycythemia, right-sided enlargement of the heart and murmur of pulmonary insufficiency; however, the characteristic feature is the striking roentgenographic evidence of a prominent pulmonary conus with large tumor-like pulsating hilar shadows which are formed by the pulmonary vessels. In the cases studied, gross examination of the heart at necropsy revealed a huge dilatation of the pulmonary artery and its main branches, a small aorta, an enormously hypertrophied right ventricle and auricle, a small left ventricle and an absence of any other congenital or acquired defect. Examination of the lungs showed that the dilatation of the pulmonary arterial tree extended peripherally almost to the pleural surface. The histologic picture of the pulmonary artery was essentially that of arteriosclerosis with intimal thickening and lipoid deposits which have not compromised the lumen. Careful histologic studies of the small arteries by Dr. Klempner revealed only slight intimal thickening—nothing to suggest the condition described by MacCallum under the title of obliterative pulmonary arteriosclerosis. Such an idiopathic dilatation of the pulmonary artery is tentatively considered here as either the result of an unequal division of the truncus arteriosus or possibly as a gigantism, an arteriomegaly, similar to the congenital arteriectasis which occurs in the limbs and has been described by Parkes-Weber as "hemangiectatic hypertrophy of the limbs."

*Dilatation of the Pulmonary Artery Associated with Cardiac Anomalies.*—A clinical and roentgenographic picture similar to the foregoing occurred in a woman of gracile habitus in whom necropsy revealed, in addition to the huge dilatation of the pulmonary artery and an occluding thrombus in each of its main branches, a large congenital interauricular septal defect. A necrotizing arteritis of the pulmonary artery affecting primarily the media was found by Dr. Klempner. The dynamics of the circulation leading to the greatly hypertrophied right ventricle and auricle are somewhat more readily understood under such conditions than in the purely idiopathic dilatation.

Still clearer from the pathologicphysiologic standpoint was another case in which the dilatation of the pulmonary artery was associated not only with an interauricular septal defect but also with an acquired mitral stenosis (Aschoff bodies were not found). Twenty-four such instances, including the one of Dressler and Roesler, have recently been collected from the literature by McGinn and White.

#### DISCUSSION

HUGO ROESLER (by invitation): This is a real contribution to the rather neglected field of pulmonary artery disease. The case of gigantism of the pulmonary artery may represent a new entity, provided that one is not dealing with

diffuse arteriovenous aneurysmal anastomoses of the arterial and venous system respectively. I am informed by Dr. Oppenheimer that careful histologic examination showed no evidence of arteriosclerotic changes. The dilatation of the pulmonary artery in the presence of an interatrial septal defect is characteristic indeed. The ratio of the size of the pulmonary artery to that of the aorta is on the average 3:2. The enormous enlargement of the heart, almost entirely on the right, with marked preponderance of dilatation over hypertrophy, can be understood only by assuming a left to right shunt, and these cases prove that the necessity of managing an increased amount of blood is the main stimulus for the dilatation. From a roentgenologic point of view it may be said that the enormous dilatation of the branches of the pulmonary artery has been misinterpreted and diagnosed as a tumor. The shadows of these vessels can be seen to pulsate, and when these pulsations become markedly expansile, regurgitation of the pulmonic valves must be present. The narrowness of the aortic shadow helps in the diagnostic approach. Another interesting feature of the interatrial septal defect is the preponderance of its occurrence in females.

I am glad that Dr. Oppenheimer has not used the term "Ayerza's disease." Confusion has been brought about by trying to construct clinical or pathologic entities and adding Ayerza's name to them. Ayerza himself (1901) described a clinical picture of what would now be called the decompensated stage of a cor pulmonale—a picture which had already been described in the French literature. The postmortem report of Ayerza's case does not mention the pulmonary artery, and Ayerza himself never made any statement as to the pathologic entity of this clinical syndrome. As to the primary arteriosclerosis of the lesser circulation, probably the best early macroscopic and microscopic description with the correct correlation of the clinical and pathologic pictures was given by J. Klob (Wechbl. d. Ztschr. d. k. k. Gesellsch. d. Aerzte, Wien 21:357, 1865). As Dr. Oppenheimer has pointed out, there are many causes for dilatation of the pulmonary artery, and I am demonstrating by lantern slides some roentgenograms illustrating the influence of thyrotoxicosis and pneumoconiosis. As another interesting contribution, I present a case of congenital heart disease with enormous dilatation of the pulmonary artery and a visible lime salt deposit in it.

As Costa has pointed out, aneurysm of the pulmonary artery as compared with aneurysm of the aorta has a rather different age distribution, as well as etiology. The former is distributed about equally throughout all age groups and occurs, in almost half of the cases, in the presence of other cardiovascular malformations, and in the rest of the cases the mycotic and infectious factors surpass the syphilitic one.

#### THE PATHOLOGY OF MEASLES WITH SPECIAL REFERENCE TO PNEUMONIA. LAWRENCE W. SMITH.

My paper concerns itself with a review of the pathologic changes found in approximately seventy-five cases of fatal measles. The cutaneous changes and the lesions of the mucous membranes are first reviewed, showing the Koplik spot to be a periductal cellular reaction and the lesions of the skin to consist of mild hyperemia with perivascular infiltration by large mononuclear cells. The chief cause of death in measles being pneumonia, a review of the pulmonary findings is presented. These may be summarized briefly as follows: There are an acute mucopurulent bronchitis and bronchiolitis associated with marked peribronchial thickening. The peribronchial thickening is caused by fibroblastic proliferation, capillary congestion with endothelial proliferation, edema and marked mononuclear cell infiltration. It is similar to the peribronchial thickening seen in pertussis and in influenza, but the type of cell involved is the large mononuclear phagocyte. From the peribronchial thickening interstitial pneumonitis develops, which seems to be nearly specific in these three diseases—measles, pertussis and influenza. This suggests the possibility of a common etiology, as emphasized by McCordock. The other visceral changes are found primarily associated with the reticulo-endothelial system and are evidenced by infiltrations of the spleen, lymph nodes and lymphoid tissue of the

gastro-intestinal tract by large mononuclear cells, with occasional instances of interstitial nephritis. Toxic changes are seen involving the central nervous system, with the development of an encephalomyopathy, as shown by Scheffer and Ferraro and by Krieder. In addition to the nerve cell degeneration, endothelial damage is seen, with venous thrombosis and similar endothelial cellular proliferation. In the treatment of the disease, no specific therapy has been discovered. Convalescent or parental serum has been used successfully as a prophylactic measure, and more recently similar results have been suggested in the use of placental extract prepared according to the method of McKhann.

#### DISCUSSION

LAWRENCE SOPHIAN: About three years ago I had occasion to examine an appendix removed from a child with measles, and at the time I was much puzzled by the appearance of certain very large cells, some multinucleated, in the lymph follicles of the appendical mucosa. About two months later Warthin's paper on the lesions of the tonsils in measles came out, and he described peculiar endothelial leukocytes or large monocytes of the same type as I saw in the appendix. I should like to ask Dr. Smith if such cells are known to occur in the lungs in the pneumonia produced in measles.

LAWRENCE W. SMITH: Yes, Dr. Sophian, they do.

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*In Conjunction with the Stated Meeting of the New York Academy of Medicine,  
April 5, 1934*

WILLIAM C. VON GLAHN, President, in the Chair

#### SYMPOSIUM ON THE RECENT PROGRESS OF RESEARCH IN LEUKEMIA

##### EXPERIMENTAL STUDIES IN LEUKEMIA. MAURICE N. RICHTER.

A general résumé is presented of the work on lymphatic leukemia of mice carried out as a cooperative project by the Department of Genetics of the Carnegie Institution of Washington and the Department of Pathology of the College of Physicians and Surgeons of Columbia University. The report concerns mainly experiments on mice of strain C58, in which about 90 per cent of the animals living more than six months show spontaneous development of lymphatic leukemia. Four types of experimental observations are described:

1. Studies on the transmissibility of leukemia. Leukemic cells from mice of strain C58 can be carried indefinitely in normal mice of the same strain by transfer from animal to animal, the inoculated animal acquiring leukemia. In order to transmit the disease by inoculation, it is necessary to introduce living leukemic cells into the bodies of susceptible mice.

2. Cytologic studies of the leukemic cells. The cells are morphologically identical with the immature lymphoid cells of the mouse, and do not present any structural abnormalities. The chromosome number is 40, which is normal for the mouse. In inoculated animals the infiltrations do not arise by proliferation of cells of the inoculated host, but by continued growth and division of the cells introduced.

3. Metabolic studies of leukemic cells. Not only are there striking differences in metabolism between the lymph nodes of normal and leukemic animals, but also between those of mice in which different lines of cells are carried by inoculation. Thus the oxygen consumption of nodes of one line (A) is greater than that of normal nodes and of leukemic nodes of another line (I). However, aerobic glycolysis of the latter line (I) is almost three times as great as that of normal nodes or of nodes of line A. Anaerobic glycolysis in line I is about 1.5 times as great as that of normal nodes.

4. Genetic studies on the occurrence and transmissibility of leukemia. Long inbreeding of strain C58 and of other strains bred in the same laboratory has led to marked genetic uniformity in each strain. The occurrence of spontaneous leukemia in a large proportion of mice of one strain and its virtual absence in another indicate that the disease is under specific genetic control. However, some non-genetic factor is also involved, as indicated by the occurrence of a larger proportion of spontaneous cases in hybrids when the mother is from the leukemic (C58) strain.

Heredity also plays a part in susceptibility to inoculation, for leukemias arising in one strain of mice may or may not be transmissible by inoculation in mice of another strain. Thus, a mouse may be susceptible to one line of leukemic cells, but not to another. These results indicate the importance of genetic control of mice used in the study of experimental leukemia.

#### EXPERIMENTAL PHASES OF LEUKEMIA. J. FURTH.

Two phases of experimental leukemia are discussed: (a) the relation of x-rays to the lymphomatosis of mice; (b) avian leukosis. The former is fully described in the *American Journal of Cancer* (19:521, 1933) and the *American Journal of Roentgenology and Radium Therapy* (in press), and the latter in the *Journal of Experimental Medicine* (58:253, 1933; 59:501, 1934) and the *Proceedings of the Society of Experimental Biology and Medicine* (31:921 and 923, 1934).

#### PATHOLOGIC ASPECT. R. H. JAFFÉ, Chicago (by invitation).

Experimental studies on leukemia in lower animals seem to lend strong support to the conception that leukemia is closely related to the malignant tumors. In human pathology, too, considerable similarities exist between leukemia and neoplasms. There is the purposeless mass production of cells of inferior biologic quality. The mitoses are atypical, resembling those seen in carcinomas and sarcomas. Isaac and Groat refer to haploid mitoses in leukemic myeloblasts. The reduction in the number of chromosomes suggests an accident in cell division resulting in the formation of viable daughter cells with abnormal growth properties and defective ability to mature. Since Banti and Ribbert, much significance has been attributed to the invasion of blood vessels by the leukemic formations.

The fact, however, that in rare cases of leukemia the leukemic process may, in one location, assume the appearance of a sarcoma with invasive, destructive growth and formation of metastases (leukosarcoma, myeloblast sarcoma) indicates that common leukemia should not be identified with malignant neoplasms. According to Naegeli and others, the long persistence of the parenchyma in organs with leukemic infiltrations (e. g., the kidneys) and the maintenance of undisturbed function by these organs are difficult to reconcile with the behavior of a malignant tumor. Under the influence of intercurrent infections (tuberculosis, erysipelas) the leukemic process may almost completely subside and the normal function of the blood-forming organs may be temporarily restored, a phenomenon not observed in the presence of malignant neoplasms. In view of these discrepancies, Helly and, to certain extent, also, Heiberg relate leukemia to adenoma rather than to a malignant neoplasm. But the invariably fatal outcome of leukemia speaks strongly against Helly's interpretation.

A close scrutiny of the histogenesis of the leukemic changes reveals, I believe, the principal differences between the neoplasm and leukemia. The sarcomas and the carcinomas start from a more or less circumscribed group of cells, and it is by unrestricted multiplication of these cells that the tumor grows and metastasizes. The majority of investigators agree on the autochthonous formation of the leukemic infiltrations, wherever they may arise, although hemorrhages may occasionally lead to the colonization of leukemic blood cells with secondary local growth (e. g., in the brain—Fried).

The question will arise as to the source of the leukemic infiltrations. Since leukemic infiltrations may occur in any organ of the body, the parenteral tissue of

these infiltrations must be widely distributed. Maximow, and with him a considerable number of recent investigators, assumes that in the mesenchyma there persists throughout life an undifferentiated germinal tissue endowed with blood cell-forming potencies. This germinal tissue, which does not store electronegative colloids or phagocytose corpuscular matter, is located between the adventitial cells of the small blood vessels. In the blood-forming organs it is inserted into the cytoplasmic reticulum. Under normal conditions the higher differentiated cells of the mesenchyma, especially the reticulo-endothelial cells (histiocytes) and the fibrocytes, do not produce blood cells. When irritated the reticulo-endothelial cells may be cast off into the blood stream as "blood histiocytes." The majority of the blood histiocytes are filtered out in the capillaries of the lung, where they disintegrate, and only a few of them reach the peripheral blood. Mass production of blood histiocytes may occasionally lead to a flooding of the peripheral blood by large phagocytosing mononuclear cells (endocarditis lenta, smallpox, malaria, kala-azar, septic reticulosis, etc.). The histologic analysis of a great number of cases of leukemia has convinced me that leukemia is characterized by the acquisition of blood cell-forming potencies by the entire mesenchyma, in particular the reticulo-endothelium and the fibrocytes. The power of differentiation into blood cells may be so strong that the reticulo-endothelial cells and fibrocytes reveal the structure of young blood cells while they are still sessile. Thus, in acute myelosis the Kupffer cells of the liver may be found filled with oxydase granules that extend into the branched processes. In a case of subacute myelosis with a predominance of eosinophilic myelocytes I have found the Kupffer cells stuffed with oxyphilic granules. In inflammatory granulation tissue in cases of myelosis I have seen neutrophilic and oxydase granules in the cytoplasm of swollen fibrocytes. In this premature differentiation of the sessile mesenchymatous cells the earliest stage of blood cell formation, namely that of the hemocytoblast, is skipped. In some cases of acute leukemia the mesenchymal cells develop into hemocytoblasts (stem cells) which enter the blood without further differentiation. These hemocytoblasts are often confused with lymphocytes.

The changes described are most striking in the cases of acute leukemia which take such a rapid course that the patient dies before the leukemic infiltrations have become too extensive to allow an analysis of the histogenesis. No matter what type of leukemia is diagnosed from the blood picture or what type of blood cell predominates in the lesions of the various organs, the leukemic formations always start in the same location.

So far as the relation between acute and chronic leukemia is concerned, I do not see any reason for considering them as two different diseases, as suggested by C. Sternberg, Gloor and others. Because of the slow development and prolonged course the histogenesis of the process is less distinct in the chronic leukemia than it is in the acute form. Because of the slow development there is also a greater difference in the distribution of the leukemic infiltrations than in the cases with rapid course. Fundamentally, however, the process is the same, as shown in those instances in which a chronic leukemia terminates in an acute one.

Recent years have brought an extensive literature on a new type of leukemia first recognized by Reschad and Schilling more than twenty years ago. I refer to the monocytic leukemia. Though questioned by outstanding hematologists like Naegeli and Carl Sternberg, there is so much evidence in favor of a monocytic leukemia that its existence can no longer be denied (Forkner, Fowler, Foord, Parsons and Butt, Clough and many others). Of thirty cases of acute and subacute leukemia which I have studied during the last three years, six were of the monocytic type. Much confusion, however, has been created by including among the cases of monocytic leukemia those with excessive, systemic proliferation of the reticulo-endothelial cells or of the reticular cells only (aleukemia, subleukemia, leukemic reticulosis or reticulo-endotheliosis). I consider these conditions as septicemias with abundant reactive proliferation of the reticulo-endothelium (see also Krahn, Akiba, Terplan and others). The appearance of numerous, sometimes phagocytosing, histiocytes in the peripheral blood does not indicate any relation to

leukemia, since the blood histiocyte is not the precursor of the monocyte, but an abnormal fully mature blood cell. The immature precursor of the monocyte is the monoblast, and it is the monoblast that characterizes the monocytic leukemia. I agree with Forkner, who traces the monoblast to the same parenteral cell from which the other blood cells are derived. Since in all types of leukemia the reticulo-endothelium acquires blood cell-forming potencies, one should not use the term "leukemic reticulosclerosis" or "leukemic reticulo-endotheliosis" as synonymous with "monocytic leukemia," as is frequently done.

A critical study of the pathology of leukemia leads to the conclusion that leukemia has just as many features in common with malignant neoplasms as it has differences from them. There are many observations pointing toward relations between leukemia and a variety of inciting injurious agents such as infections, poisons (benzene), roentgen rays, trauma and dietary deficiencies. These observations are too numerous to be merely incidental. On the other hand, it is only in rare instances that a leukemia develops following the causes quoted. Hence one is compelled to assume disposing congenital factors, an abnormal constitution of the mesenchyme. In this connection reports on the familial occurrence of leukemia are of great interest (Weiss and Thums, Brugger, Wolff, Vercelotti, Riccitelli, Sorrentino, S. Petri, V. Deutsch, Morawitz and others). For these reasons it appears advisable not to identify leukemia with malignant neoplasms, but to consider it as a definite and separate disease entity.

#### CLINICAL AND THERAPEUTIC ASPECTS. LLOYD F. CRAVER (by invitation).

Despite extensive clinical observations of leukemia in recent years, there is nothing essentially new in its treatment. These observations have been of value chiefly in bettering one's understanding of the scope of the disease and in improving the technic of treatment.

Emile Weil and Isch-Wall's statement that the appearance of gross leukemic infiltrations, which they term "tumors," of skin, bone or external lymph nodes in the course of chronic myeloid leukemia signifies the terminal stage is true in general, but there are exceptions.

Priapism is much more rare than is generally believed. In a series of over a hundred male patients with leukemias observed at Memorial Hospital it occurred in only one.

In recent years several authors have discussed the interesting question of the relationship between erythremia and leukemia. Parkes-Weber speaks of erythro-leukemia.

Infectious mononucleosis is frequently mistaken for lymphatic leukemia. Bunnell's report shows that the heterophile antibody reaction may be of great use in the differential diagnosis of infectious mononucleosis. However, the worth of this test cannot be judged without further extensive trials. It seems possible that what produces infectious mononucleosis in one person may cause leukemia in another.

Splenocontraction induced by intramuscular injection of epinephrine hydrochloride is believed by some French workers to be of value in certain doubtful cases, as judged by the effect produced on the blood count.

In treatment, more particularly of myeloid leukemia, the nearer the white cell and differential counts are brought toward normal, the better and more lasting are the results.

Forkner's use of "relentless" doses of arsenic has served to recall attention to this agent.

Nemenow suggested preliminary irradiation of the kidneys so as to free them from leukemic deposits and thus lessen the retention of uric acid. Arendt and Gloor suggested diathermic treatment of the kidneys in order to increase the circulation of blood through them and thus increase their eliminative function.

Friedgood drew an interesting parallel between exophthalmic goiter and chronic lymphatic leukemia, and has tried using iodine in lymphatic leukemia, with some palliative results.

The value of transfusions in leukemia is questionable.

The hemorrhagic diathesis has been lessened in some cases by exposure to mild actinic rays, together with a high vitamin intake.

Infected wounds may be very troublesome in leukemic subjects. In several cases rapid healing has resulted from small doses, from 100 to 200 roentgens, of lightly filtered, low voltage x-rays.

The infiltrations that rather commonly appear about the eyelids or orbits in lymphatic leukemia may yield readily to small doses of x-rays.

*Total Irradiation.*—For a number of years certain clinicians have treated leukemia by irradiation of the entire body. Their methods have been two: (1) to remove the roentgen tube a sufficient distance from the patient's body so that the beam of x-rays will include all or the greater part of the body; (2) to expose in succession large fields until the entire body has been treated. In general, the results of total irradiation have been about on a par with those of the usual methods of local irradiation; sometimes the results in the blood count and the size of the spleen have been less satisfactory.

Beginning in May 1931 and continuing for two years, my associates and I tested a new method of total irradiation, known as the Heublein method. Patients were treated continuously, day and night, with very weak intensities of radiation from a tube operating at 185 kilovolts placed at a distance of from 5.4 to 7.3 meters from the patients. By this method we treated five persons with myeloid leukemia, twenty-seven with chronic lymphatic leukemia and nine with so-called lymphatic pseudoleukemia. Of these forty-one patients fifteen showed improvement, and up to August 1, 1933, were living 4.5 to 21 months after treatment was begun. Ten showed distinct palliation but succumbed to the disease in from 3.5 months to 2 years after treatment. Sixteen patients failed to show improvement. However, we were experimenting with an entirely new method, and we feel that the poor results were largely accounted for by errors in the dosage and in the selection of cases. There is no doubt that in many of the cases, particularly in those of the lymphatic group, the treatment gave results that were at least equal if not superior to those obtainable by local irradiation. Our impression is that myeloid leukemia does not do so well as lymphatic leukemia following total irradiation, although we treated only five patients with myeloid leukemia.

#### DISCUSSION

FRANCIS CARTER WOOD: I have been treating patients with leukemia for thirty years by a combination of x-rays, radium, arsenic and similar drugs without, it seems to me, very much improvement in the results. These patients may be divided into two classes: First, those with acute leukemia, which tends to become hemorrhagic, in whom as a rule no beneficial result comes from any type of treatment. They are made worse by irradiation; they are not regularly benefited by transfusion; arsenic is useless, and most of them die within a short time. There are exceptions, of course. I have seen a patient symptomatically cured for a number of months by transfusions, but the disease returned in an even more acute form. The second type are the patients with chronic leukemia, and of these the ones with the lymphatic type of leukemia may be divided into three groups radiologically: a small group who, with suitable doses of x-rays applied to the spleen and nodes, will live for a period of years. They are usually older patients, and in them conditions are very favorable. I have had a number live for six to nine years with very little x-ray therapy, just enough to keep the count down and to keep the nodes from causing discomfort. There is another type in whom the disease is fairly rapidly progressive, and in whom irradiation does very little, except to reduce the size of the spleen and nodes, but they die without any particular benefit. Between these lies a group in whom palliation for a year or two is often obtained. In the type with acute myelogenous leukemia no benefit is obtained. In the type with chronic myelogenous leukemia, despite the statement that no prolongation of life is obtained, an amelioration can be brought about with x-rays in many

cases, and when one sees a practically moribund patient get up and go back to work for two or three years it is difficult to understand why one should think life cannot be prolonged.

It is difficult to classify these patients. They vary greatly under different types of treatment, no matter whether arsenic, which Naegeli has strongly recommended lately, or x-rays, which most favor, are used. Most physicians do not like to get the leukocytes down to normal, because every once in a while they go far below normal, and the patient dies with pneumonia or some other infection.

It is wise to keep the patient thoroughly informed as to the dangers of even minor surgical operations and pulling of teeth, the latter often setting up an intractable hemorrhage which may lead to the death of the patient in days or weeks.

I have succeeded in getting two women through pregnancy by careful irradiation up to the time of delivery, but in young women irradiation sterilization should be done.

Benzene is a very poor drug. It often causes a hemorrhagic diathesis and multiple lesions of the skin, and is not employed in my clinic in the treatment of leukemia. Recently Kraemer of Philadelphia has tried the use of lead, and has reported good results in a few cases, but such reports have no general value, because patients vary, and each must be treated cautiously and individually. However, there may be something in this treatment in that the lead is phagocytosed and carried to the spleen and bone marrow, and possibly has a toxic effect on the cells, so that the amount of x-rays necessary is diminished.

NATHAN CHANDLER Foot: You have seen and heard the evidence on both sides of the question for and against leukemia as a tumor, and you realize where the poor pedagogues stand in regard to this matter when it comes to teaching students what the condition may be. One might think of the disease as a scale or a gamut, and range at one end those conditions in which the blood stream is flooded with the atypical white cells and the blood-forming organs are abnormally active, and at the other end of the scale those definitely localized myeloblastic tumors which are disseminated very much like ordinary tumors insofar as they metastasize widely (chiefly to the bone) and fail to flood the circulation with white cells. In the middle of this scale would come those types in which both of these observations are made: the circulation is flooded, and subsidiary tumors or infiltrations are formed. I think these daughter growths are not always so well borne as Dr. Jaff's reference to Naegeli would indicate, for, particularly in chloroma in which one sees subsidiary tumors of a green color throughout the body, a good deal of damage to tissue results, and this is also indicated in what has been said concerning the kidneys.

One might make a similar scale in connection with leukemias of the lymphoid type, and then from this pass on quite naturally to those of the monocytic type. The chief stumbling block in the attempt to draw analogies between malignant tumors and the leukemias is this: One is trying to compare two pictures that are, from the histologic and physiologic standpoint, so fundamentally different under normal conditions that one is bound to fail if one pursues the attempt too closely. The blood is the most ubiquitous and labile of all the body tissues. One can get along fairly well with the analogy up to a certain point, but the normal and rapid circulation of the blood as a tissue is without analogy, and therefore a tumor of such a tissue is in a class by itself. Malignant tumors may use the blood stream as a means of transportation for their daughter cells, they themselves remaining fixed; in the leukemias, the tumor is an integral part of the moving vehicle itself; therefore if one merely attempts to adjust one's conception of malignant tumors of fixed tissues to the conception of a tumor in which the component cells are constantly being shuffled in a shifting and moving tissue, one will find the comparison not so fantastic as one at first conceived it to be.

That the cells of such a tumor may arise anywhere in the body as a development of Maximow's polyblasts may or may not be a fact; it still is under lively dispute, and, alluring as this theory may be, as one grows older and more cautious, one is less inclined to accept it unless its validity is conclusively proved by obser-

vations in vivo. The mere presence of oxyphilic or other granules common to granulocytes in cells that are notoriously avid phagocytes cannot be accepted as proof of their transformation into granulocytes. The theory is well worth investigating, and one must strive to maintain an open and unbiased attitude toward such questions. Maximow maintained that there is a supply of polyblasts throughout the body, cells that are mesenchymal in origin and that never reach a fixed condition of differentiation. They constitute, so to speak, a reserve force of persisting embryonal tissue that has the capability of becoming differentiated into a number of other forms, such as blood cells, if the occasion arises. They may form histiocytes, fibroblasts, lymphocytes and so on, and under certain circumstances, according to Maximow, the process may be reversed, the differentiated cell undergoing dedifferentiation and returning to the potential embryonal condition of the polyblast. Such a theory, while it is applicable within limits to inflammation and repair, becomes absurd if it is pursued to its logical conclusion. Mesodermal tissue can also produce epithelium, and as a result of this one finds some authors bold enough to derive epithelial tumors from these polyblasts and to explain the occurrence of cancer on such a basis.

Dr. Jaffé strikes a sympathetic note when he excludes the so-called aleukemic reticulosis from the leukemias. In a recent paper read before the New York Pathological Society I reported a case of that disorder and took the same stand on exactly the same grounds. To both of us the maturity of the monocytes in contrast with the immaturity of the leukemic cells and the almost invariable history of infectious disorders in connection with this disease prior to its development seemed very much against its being a leukemia. There are conditions in which the mononuclear cells may assume embryonal characteristics, such as generalized reticulosarcomatosis, and these are very different from the aleukemic reticulosis that Dr. Jaffé has mentioned.

This leads to the theory that infection is the cause of leukemia. Infections do call out large numbers of polymorphonuclear leukocytes in cases of ordinary leukocytosis. Many of these leukocytes may be of an immature type, a fact of some importance to the hematologist. One may see histologic pictures closely resembling leukemia of the myeloid type in the tissues of children who have died from acute infectious diseases, such as diphtheria and scarlet fever, with extensive myelocytic and even myeloblastic infiltrations of various organs. Ignorant of the clinical histories in such cases, one might be misled to diagnose the condition as myeloid leukemia, which would be quite erroneous.

The popularity of the old terms "acute leukemia" and "chronic leukemia" is bound to wane the more investigators work along the experimental pathways followed by Dr. Richter and Dr. Furth, for the more one knows about this disorder the less one is inclined to regard it as an inflammation to which the terms "subacute," "chronic" or "acute" may be applied. It is rather a question of the degree of malignancy as one sees malignancy illustrated in carcinomas—it may wax or wane in leukemia just as it may in neoplasms. Although I am not an enthusiast for the grading of tumors, one might grade the leukemias in some such fashion, having grades 1, 2 and 3 according to the rapidity of the growth, the extensiveness of the infiltration and so on.

NATHAN ROSENTHAL: Leukemia presents marked variations with respect to the symptoms, course and blood picture. The symptoms alone cannot be relied on for the diagnosis, which must rest on the characteristic blood changes. These do not depend on the number of white blood cells so much as on the presence and persistence of specific types of cells, such as myelocytes, myeloblasts and a relative and absolute lymphocytosis and monocytosis. It is interesting to note that the frequencies of the three main types of leukemia—myelogenous, lymphoid and monocytic—correspond with the percentages of the three main types of leukocytes in the circulating blood. The underlying systemic disorders are essentially the same in all cases, and their neoplastic or malignant character is evident. Further arbitrary divisions of the cases can be made according to the duration of the disease, acute or chronic, and according to the number of white blood cells, mainly

leukopenic or leukocytemic. In other words, the white blood cells in leukemia may vary from 400 to 1,000,000 in number.

The transmission of the disease in animals is an interesting problem. So far there is no information with respect to its transmission in man. I have observed this possibility in two patients as the result of blood transfusions from patients with polycythemia to patients with severe anemia in whom the leukocytes were not abnormal. In both patients, about a year after many transfusions were given, acute myeloblastic leukemia developed. This may indicate the close relationship of polycythemia and leukemia. In fact, leukemia is not infrequently the end-stage of polycythemia.

The treatment of leukemia is largely symptomatic. Arsenic, transfusions and particularly roentgen ray irradiation are the chief means of inducing symptomatic improvement, remission or possibly prolongation of life. Persons with acute leukemia may also be irradiated. In two such patients remissions were induced similar to those obtained in patients with chronic leukemia. No ill effects have been noted in the treatment of a large number of patients with acute leukemia.

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## PHILADELPHIA PATHOLOGICAL SOCIETY

*Regular Meeting, March 8, 1934*

MORTON McCUTCHEON, *President, in the Chair*

TENTH-NORMAL HYDROCHLORIC ACID AS A DILUTING FLUID FOR COMBINED LEUKOCYTE AND HEMOGLOBIN DETERMINATIONS. C. A. PONS and WILLIAM P. BELK.

With the use of the Haden-Hausser hemoglobinometer, for which a 1:20 dilution of blood in tenth-normal hydrochloric acid is made in a white cell pipet, an economy of apparatus and time would result if the white cells could be determined on the same preparation. Fifty-eight duplicate counts were made with tenth-normal hydrochloric acid and 0.5 per cent acetic acid. Counts on leukocytosis and lymphocytosis as well as on normal blood were included. In the hydrochloric acid preparations there was a slightly brownish background and the cells were smaller than in acetic acid. These differences were not sufficient to interfere with accurate counting. The average deviation of the two series was 6.09 per cent, the counts on the hydrochloric acid preparations being somewhat lower. However, when ten hydrochloric acid preparations were recounted after an interval of two hours, the second counts averaged 8.7 per cent higher than the first ones. This indicates that the white cells are preserved in this diluent for that period of time. A few observations on the distribution of the cells in the counting chamber with both diluents indicate that this factor alone may account for such differences as appear in the two series. Granted that a 10 per cent variation is permissible in the enumeration of white cells, tenth-normal hydrochloric acid appears to give accurate results.

EXPERIENCES IN EVALUATION OF THE GRUSKIN SKIN TEST FOR CANCER.  
JOSEPH MCFARLAND, M. FRIEDMAN and J. H. CLARK.

The Gruskin skin test for cancer was performed on 174 persons in the Philadelphia General Hospital. The antigen, prepared from the liver of fetal calves, was supplied either by Dr. Gruskin or by Sharpe and Dohme, Philadelphia, who prepared it under his direction. In all, 17 antigens were used. The patients tested were divided into two groups. Group 1 (controls) totaled 71 persons; 13 were presumably normal nurses or physicians; 48 presented 24 common diseases, not malignant; 10 complained of tumors, not cancerous (fibro-adenoma, Hodgkin's disease, giant cell tumors, osteoma, etc.). In this group 85 tests were performed,

yielding negative results in 74.1 per cent. Group 2 included 103 patients suffering from cancer, diagnosed by biopsy in 57 instances and clinically in 44; 114 tests were performed. The results were as follows:

	Cases	Results of Gruskin Tests		Tests	Per Cent Positive
		Positive	Negative		
Carcinoma (diagnosed by biopsy)					
Irradiated.....	32	28	6	34	82.3
Nonirradiated.....	27	22	10	32	68.7
Carcinoma (diagnosed clinically by x-ray picture, etc.)					
Irradiated.....	24	18	8	26	69.2
Nonirradiated.....	20	18	4	22	81.8
Totals.....	103	86	28	114	75.4

The results of the total series of tests were as follows:

	Cases	Tests	Results	Per Cent Accurate
Controls.....	71	85	Negative 63	74.1
Carcinomas.....	103	114	Positive 86	75.4
Totals.....	174	199	149	74.8

Our impressions of the test are: 1. It is based on a false immunologic premise; i. e., that an autochthonous growth of cells, possibly fetal in origin, certainly human in type ontogenetically, will produce a sensitivity in the fixed tissue cells to a phylogenetically foreign protein of embryonal type. 2. It is not and probably cannot be sufficiently well standardized to prevent conflicting results. 3. The reactions are indefinite and extremely hard to read. 4. The reactions are not typical of true allergy. 5. In no case was a cancer diagnosed by the test that could not have been recognized by more simple means.

#### THE RELATIONSHIP BETWEEN RHEUMATIC AND SUBACUTE BACTERIAL ENDOCARDITIS. WILLIAM C. VON GLAHN and ALWIN M. PAPPENHEIMER, Columbia University.

A series of twenty-six cases of subacute bacterial endocarditis was studied in order to make clear the relation between the underlying rheumatic disease and the bacterial infection. Histologic evidence of coincident active rheumatic lesions in the form of fresh verrucae was found in every case. In several instances, rheumatic vegetations free from demonstrable bacteria were present on valves unaffected by the bacterial infection. The incidence of Aschoff bodies in the myocardium was the same as in cases of uncomplicated rheumatic carditis. The more plausible inference from these observations is that, in the rheumatic subject, the subacute bacterial infection of the valves is implanted on active rheumatic vegetations.

#### RETOTHelial (RETICULO-ENDOTHELIAL) SARCOMA. R. PHILIP CUSTER.

Six cases of malignant tumors arising in cells of the reticulo-endothelial system are presented. From this material and published cases of others the following points are emphasized:

The predominant incidence is in the fifth and sixth decades of life.

The characteristic clinical features are: regional pain, a febrile course and moderate anemia, with progressive loss of weight and terminal cachexia.

The predominant site of origin is in the lymph nodes or the spleen. An occasional primary focus is in the pharynx or along the gastro-intestinal tract.

Propagation is by direct extension, lymphatic permeation and/or blood stream metastasis.

The duration is relatively short; in the group studied it was from five to twenty months following the onset of symptoms. The duration of one case was forty-

one months, but the sarcomatous condition was preceded by intestinal tuberculosis (demonstrated by laparotomy), and the onset of the tumor was indefinite.

The histologic criteria for diagnosis are: polymorphism of cells with morphologic evidence of their histiocytic nature, i. e., clear vesicular nuclei, abundant cytoplasm with vacuolation, delicate processes and phagocytic quality, and tendency to form mononucleated and multinucleated giant cells; occasional appearance of myeloid metaplasia within the tumor, and, finally, transitional forms between reticular and endothelial cells and tumor cells, the latter demonstrable in early cases before the entire architecture of the lymph node is lost.

The differentiation from lymphosarcoma and from multiform spongioblastoma is based on clinical and histologic grounds. Differentiation must be made from leukemic and aleukemic reticulosclerosis (reticulo-endotheliosis).

It is recommended that pathologists adopt the term "retothelial sarcoma" (*Retrothelsarkom*—Roulet) to serve for the terms "reticulum-cell sarcoma," "reticulum-cell lymphosarcoma" and "Hodgkin's sarcoma."

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*Regular Meeting, April 12, 1934*

MORTON McCUTCHEON, *President, in the Chair*

The William Wood Gerhard Gold Medal of the society was presented to Dr. George H. Whipple, after which Dr. Whipple gave the Annual Conversational Lecture, his subject being: "Regeneration of Hemoglobin and of Blood Plasma Proteins Controlled by Diet Factors."

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*Regular Meeting, May 10, 1934*

MORTON McCUTCHEON, *President, in the Chair*

CERTAIN FEATURES OF ARTERIOSCLEROSIS IN WILD ANIMALS. HERBERT FOX.

It has been possible by a combination of the Gömöri silver photographic method and the Spalteholz translucency method to obtain specimens in which a black precipitate is deposited in the areas that appear to contain calcium when sectioned and stained by hematoxylin and alizarin. Attention is directed to the provisions by Gömöri that this silver precipitation may not penetrate the entire mass but be limited to a narrow rim around such degenerations.

Incineration of arteries by the Policard method has added little to knowledge of the anatomy of vessels and the lesions of arteriosclerosis. Fibrillary thickening of the intima is accompanied by a striated ash. Atheroma of the intima shows a finely stippled ash. The ash of the media follows the lines of the elastica. The ash of the avian intima is coarser than that of the mammalian; this is consistent with the character of the elastica as shown by Weigert's stain in the two kinds of vessels. Heavy calcification shows a brilliant silver ash.

- One feature of comparative arteriosclerosis may assist in forming an opinion about Thoma's theory in certain cases of bovine and psittacine arteriosclerosis: There is definite evidence that muscle degenerates before elastica, and that the elastic tissue and the whole wall may give way; the thickening of the intima may be for the purpose of retaining the lumen.

THE RESULTS OF THE STUDY OF SWINE INFLUENZA AND THEIR POSSIBLE APPLICATION TO THE HUMAN DISEASE. RICHARD E. SHOPE, Rockefeller Institute, Princeton, N. J.

Swine influenza is a disease which supposedly appeared for the first time in the fall of 1918 coincident with the great pandemic of human influenza. The disease has a sudden onset and is highly contagious. The salient clinical features

are fever, anorexia, extreme prostration, cough, a rapid diaphragmatic type of respiration and leukopenia. The mortality ranges from 1 to 15 per cent, death or recovery occurring after from two to six days of illness. The disease is readily transmissible from pig to pig either by contact or by intranasal inoculation with suspensions of infected lung or exudate from the respiratory tract of a sick animal.

The pathologic picture seen in animals killed on the second to the fourth day of illness is essentially that of an exudative bronchitis and massive lobar or lobular pulmonary atelectasis. The bronchi and bronchioles are filled with an exudate of polymorphonuclear leukocytes; the bronchial epithelium is fragmented or desquamated, and there is a peribronchial round cell infiltration. The alveolar walls are wrinkled, thickened and infiltrated by round cells. In fatal cases there frequently is an intensely edematous type of pneumonia.

The disease has been shown to be caused by the combined action of a filtrable virus and a hemoglobinophilic bacterium, *Haemophilus influenzae-suis*. Animals infected with the virus alone show an extremely mild and scarcely recognizable illness. Animals inoculated intranasally with pure cultures of *H. influenzae-suis* do not become ill and show no pathologic alteration at autopsy.

A comparison of what is known concerning swine influenza with the better known facts about human epidemic influenza indicates certain suggestive analogies between the two diseases. Taking into account differences in the anatomic and physiologic make-up of hogs and man, the clinical picture of the swine disease is strikingly similar to that of influenza in man; pathologically, fatal cases of the swine disease are strongly reminiscent of corresponding cases of the human disease. A hemoglobinophilic bacterium is prominently associated with both, and the organism encountered in the swine disease could not regularly be distinguished with certainty from that found associated with the human disease. The swine organism, *H. influenzae-suis*, is known to be etiologically essential to the production of the disease; the rôle played by the human organism, *H. influenzae*, is highly controversial.

Recently Smith, Andrewes and Laidlaw (Smith, Wilson; Andrewes, C. H., and Laidlaw, P. P.: *Lancet* 2:66, 1933) obtained from persons with influenza a virus that is pathogenic for ferrets. Indirect experimental evidence strongly suggests that this virus is of etiologic significance. They furthermore observed that the swine influenza virus was also pathogenic for ferrets and produced a disease in these animals that was not only similar to that produced by the human virus but appeared to confer some cross-immunity to the human virus.

Since in swine influenza both the filtrable virus and *H. influenzae-suis* are known to be etiologically essential, an interesting possibility, made very apparent in considering features of similarity between swine and human influenza, is that the human disease may likewise have a two factor etiology: the virus of Smith, Andrewes and Laidlaw and *H. influenzae*.

## Books Received

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DIE HORMONFORSCHUNG UND IHRE METHODEN. Max Reiss, Dr. Med., Dr. rer. nat., Privatdozent für pathologische Physiologie an der Deutschen Universität in Prag. Price, 15 marks. Pp. 415, with 26 text figures. Berlin and Vienna: Urban & Schwarzenberg, 1934.

ANNALS OF THE PICKETT-THOMSON RESEARCH LABORATORY. VOLUME X. INFLUENZA (PART II), WITH SPECIAL REFERENCE TO THE COMPLICATIONS AND SEQUELAE, BACTERIOLOGY OF INFLUENZAL PNEUMONIA, PATHOLOGY, EPIDEMIOLOGICAL DATA, PREVENTION AND TREATMENT. D. and R. Thomson. Price, \$17.50. Pp. 1557. Baltimore: Williams & Wilkins Company, 1934.

PROCEEDINGS OF THE PATHOLOGICAL SOCIETY OF PHILADELPHIA. NEW SERIES, VOLUME XXXI; OLD SERIES, VOLUME XLIX. Containing the transactions of the Society from January 1931 to January 1934. Edited by Herbert L. Ratcliffe, Sc.D., Secretary-Treasurer and Recorder. Philadelphia, 1934.

ARBEITEN AUS DEM SERO-BAKTERIOLOGISCHEN INSTITUT DER UNIVERSITÄT HELSINKI (HELSINGFORS). Herausgegeben von Prof. Dr. Osw. Streng. Band VI (1933-1934). Helsingfors, 1934.

AMERICAN TYPE CULTURE COLLECTION CATALOGUE OF CULTURES, 1934. Edition 3. Pp. 80. Chicago: John McCormick Institute for Infectious Diseases, 1934.

BIDRAG TIL SPØRGSMÅLET OM RELATIONEN MELLEM B-VITAMINERNE OG ERNAERINGENS INHOLD AF PROTEIN, FEDT OG KULHYDRAT. WITH AN ENGLISH SUMMARY. P. Vogt-Møller, Reservelag ved St. Elisabeth's Hospitals Medicinske Afdeling, København. Pp. 165. Copenhagen: Levin & Munksgaard, 1934.

PHYSIOLOGY IN HEALTH AND DISEASE. Carl J. Wiggers, M.D., Professor of Physiology in the Western Reserve University School of Medicine, Cleveland. Price, \$9. Pp. 1184, with 182 engravings. Philadelphia: Lea & Febiger, 1934.

AMEBIASIS AND AMEBIC DYSENTERY. Charles F. Craig, M.D., M.A. (Hon. Yale), F.A.C.P., F.A.C.S., Colonel, United States Army, retired; D.S.M.; Professor of Tropical Medicine and Head of the Department of Tropical Medicine, Tulane University of Louisiana School of Medicine, New Orleans; formerly Commandant, Army Medical School, and Director of the Department of Clinical Pathology and Preventive Medicine and Assistant Commandant, Army Medical Center, Washington, D. C. Price, \$5. Pp. 315, with 54 illustrations. Springfield, Ill.: Charles C. Thomas, Publisher, 1934.

## Book Reviews

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**Diseases Peculiar to Civilized Man: Clinical Management and Surgical Treatment.** By George Crile, M.D. Edited by Amy Rowland. Price, \$5. Pp. 427, with 41 illustrations. New York: The Macmillan Company, 1934.

This book is based on the assumption that so-called neurocirculatory asthenia, hyperthyroidism, peptic ulcer, diabetes and epilepsy are related diseases. These diseases are assumed to result from disturbances of the glandular and autonomic nervous systems caused by the "tension" of highly civilized life.

The first part is devoted to the elaboration of the fundamental assumption that the diseases just mentioned are the direct outcome of civilized life. Arguments are advanced from the fields of phylogeny and ontogeny. The hypothesis of orthogenesis is invoked. According to this hypothesis, a tendency of protoplasm to develop in a certain direction, once set, cannot be arrested, but may continue to the great injury and even extinction of the species. The extinction of certain dinosaurs has been explained according to this notion as caused by the great size of the body and the weight of the armor, and the Irish elk is assumed to have been destroyed through the overgrowth of its antlers. And now the human species is assumed to be on the way to "hyperkineticism," due to an excessive evolution of the energy-transforming system which includes the brain, the thyroid, the suprarenals and the sympathetic nerves. The "kinetic diseases"—neurocirculatory asthenia, hyperthyroidism, peptic ulcer, etc.—are assumed to be the outcome of the racial physiology peculiar to the rise of man in civilization and to affect especially persons of outstanding intellectual and emotional qualities. In the course of the discussion of these assumptions statements are made that arouse doubt. Here is one: African natives do not have peptic ulcers, nor do morons, because they are sheltered from the worries of civilized man. It is also suggested without adequate basis that the diseases in question, at least in part, are of comparatively recent origin. In fact, is the assumption that there is a hyperdevelopment of the "energy system" peculiar to civilized man justified? The author does not hesitate to follow his assumptions confidently to the limit: "Since all parts of the brain-thyroid-adrenal-sympathetic system are involved in the hyperactivity which produces these diseases [neurocirculatory asthenia, etc.], it is obvious that treatment may include only one point of attack or more than one, the objective in each case being to lessen the factors that are causing the damaging kinetic drive. Thus they may be treated by lessening the driving power of the brain by rationalization, or by excision of the sympathetic ganglia; by lessening the driving power of the thyroid gland by thyroidectomy or by interference with its sympathetic nerve supply by ligations; by lessening the driving power of the adrenals by denervation; or in certain cases certain of these procedures may be combined."

The second part describes in some detail "the diseases of civilized man," particularly so-called neurocirculatory asthenia, which, in spite of all that is stated about it, does not stand forth as a definite and distinct clinical or etiologic entity. The third part deals with matters of surgical technic, and the fourth part, the longest, contains histories of cases. Finally comes a general summary.

In the part relating to treatment the main emphasis is laid on operations on the suprarenal, the "brain" of the sympathetic system. Eventual suprarenal denervation became the routine treatment for patients with so-called neurocirculatory asthenia and "has been used successfully in the treatment of recurrent hyperthyroidism and of certain cases of primary hyperthyroidism, in the treatment of a limited series of cases of diabetes." The operation has proved of value also in certain cases of epilepsy. But the evidence is not in any sense convincingly in favor of the operation. There are no series of control cases. The influence of other factors besides the operation is not considered. No study has been made of the

structural changes following the operation. Is the "cut in the lines of communication" permanent? There is, in fact, no evidence adduced to show that suprarenal denervation interferes with suprarenal function.

This is a book of unfounded and unverifiable assumptions. That human beings are experiencing hyperkineticism from the excessive evolution of the energy-transforming system is an assumption. That so-called neurocirculatory asthenia, hyperthyroidism, peptic ulcer, diabetes and epilepsy are caused by hyperactivity of the kinetic system is also an assumption. That suprarenal denervation or other operations assumed to be "dekineticizing" cure these diseases or any of them is another assumption.

No consideration is given to the constant tendency in nature toward balance or equilibrium or to Cannon's conception of homeostasis, which sets forth the idea that any tendency toward change is met by increased effectiveness of the factor or factors which resist the change.

**Die pathologisch-anatomischen Grundlagen der Chirurgie des Rektumkarzinoms.** By Priv.-Doz. Dr. Heinrich Westhues, erster Oberarzt der chirurgischen Universitäts-Klinik, Erlangen. With an introduction by Prof. Dr. Schmieden, Frankfort-on-Main. Price, 29.50 marks. Pp. 113, with 107 illustrations. Leipzig: Georg Thieme, 1934.

This small book of 113 pages is a valuable and authoritative contribution. It is in large part a study of rectal polyps and their relation to cancer. It is liberally illustrated with reproductions of specimens and photomicrographs, many of which are in color; a full bibliography is appended.

The author has had the opportunity to investigate much clinical material, and judging from his description of his gross specimens he has been painstaking in his investigations. He approaches the subject as a surgeon with sound basic training; more than three fourths of the book is devoted to the pathologic anatomy of the disease. He classifies rectal polyps into three groups, as has been done by others, but his classification is his own. It is based on the degree of cell differentiation and the general structure of the gland tracts. He emphasizes the clinical application of this classification. In group 1 are the purely benign polyps. In group 2 are those in which a progressively spreading epithelial variation occurs; while most grow to a good size without malignant changes, they eventually become malignant in a large number of cases. Group 3 is made up of growths with marked atypical epithelial changes, many of which are carcinomatous. Pea-sized carcinomatous polyps are not infrequent. The book is divided into three sections: first, that on the structure of rectal polyps; second, that on the growth and local distribution of carcinoma of the rectum, and third, that on the clinical application of the study. In the first section the author states: "I determined that in about 45 per cent of all rectal carcinomas, the origin from polyps is highly probable. Together with a previously mentioned 15 per cent of absolutely certain cases, we can state, therefore, that about 60 per cent of all cases can be proved or determined as highly probably of polyp origin." In the second section it is pointed out that local metastases occur from below upward, that they are practically never below the rectal growth (excluding, of course, anal carcinoma). In the last section it is emphasized that rectal fixation in itself is not a contraindication to operation and that conclusions as to either local metastases or metatases to the liver cannot be drawn from the size of the growth. The demands of complete (radical) resection of the rectum for cancer are fulfilled by excision of the bowel 2 cm. above and below the growth and removal of all perirectal connective tissue and fat at its level and from 10 to 12 cm. above it. If polyps are present, as they often are, the polyp-bearing area is excised, if this can be safely done. The author emphasizes again that most rectal cancers develop from polyps and that the majority of polyps become malignant. The book merits the attention of pathologists and is a worthy guide to the surgeon.

**A Textbook of Histology.** By Harvey Ernest Jordan, A.M., Ph.D., Professor of Histology and Embryology, University of Virginia. Sixth edition. Cloth. Price, \$7.50. Pp. 738, with 610 illustrations. New York & London: D. Appleton-Century Company, Inc., 1934.

This is a standard and well established textbook of histology. The first edition was published in 1916. In this edition the text has been revised and in part rewritten. The sections that have received most attention in the revision are, according to the preface, those dealing with the blood, the reticulo-endothelial system, the endocrine glands, the striped muscles, the neuroglia, the nerve tissue, the reproductive organs and the lymphoid organs. However, the book has not been increased in size over that of previous editions. New illustrations have been introduced, and the book is suitably and richly illustrated. The style is clear and orderly. The functions of various structures are described briefly. The work merits the popularity that it has attained.

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## APPOINTMENTS

**INDIAN RESEARCH FUND ASSOCIATION—AP-**  
plications are invited from experts on nutrition to undertake independent charge of nutritional research under the Indian Research Fund Association at Coonoor, a hill station (6000 ft. above sea level) in the Madras Presidency of India. 2. Candidates must be graduates in medicine who have a wide experience of nutritional research both in the field and in the laboratory, and who have made original contributions on the subject; they must be of sound constitution and not more than 45 years of age; candidates must possess a sound knowledge of English. 3. Pay in the scale of Rs. 1250-100-1750 with usual departmental travelling and halting allowances; in addition an overseas pay of Rs. 500/- per mensem will be given to a person of non-Asiatic domicile, if appointed; the commencing pay of the selected candidate may be fixed at a higher rate than the minimum of the scale of Rs. 1250-100-1750 if the experience and qualifications of the candidate selected justify this; at the current rate of exchange \$1 is approximately equal to 2 $\frac{1}{2}$  Rupees; the commencing rate of pay plus overseas pay is approximately equal to \$656 a month. 4. The appointment will be for three years in the first instance, renewable thereafter. 5. An officer recruited out of India will be entitled to free passages as shown below: (i) if of non-Asiatic domicile first class 'C' by P. & O. or equivalent to India and return passage of the same class on termination of his appointment; (ii) if of Asiatic domicile, first class 'C' by P. & O. or equivalent to India; (iii) free passages as above will also be provided for wife, if married. 6. Leave according to the rules of the Association. 7. All applications must be made on the prescribed form copies of which can be had from the High Commissioner for India, India House, Aldwych, London, W.C.2. 8. Final date for receipt of completed forms of application is 30th November, 1934.

# ARCHIVES OF PATHOLOGY

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## PLASTIC STUDIES IN ABNORMAL RENAL ARCHITECTURE

### II. THE MORPHOLOGY OF THE ABNORMAL NEPHRON IN TERMINAL HEMORRHAGIC BRIGHT'S DISEASE

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AND

ANN SEAWARD LUEY, A.B.

BROOKLYN

In the first study of this series two types of the architectural unit from the kidney in the terminal stage of hemorrhagic Bright's disease were presented in plastic three dimensional form by means of models reconstructed by the Born wax plate method.<sup>1</sup> The need of such an objective representation of so complex a morphologic structure as the abnormal renal unit is manifest, but while a casual glance at the complicated pattern of the histologic section shows at once that an infinite variety of modifications of these two simpler types must exist, actual experience with the laborious procedure of model building soon convinces one that a knowledge of their diversity can never be obtained by its time-consuming employment. Fortunately an auxiliary method is at hand, which, though it lacks certain of the advantages of the presentation by models, furnishes us with the ability to study in a relatively short time a large number of units in a three dimensional form that can, if desired, be registered stereoscopically. Not only are the multi-form varieties of alteration thus available for consideration, but quantitative studies become possible so that the significance of any particular change can be determined by the frequency of its occurrence. This is the method of maceration and microdissection, the method by which Huber<sup>2</sup> and Peter<sup>3</sup> established in its modern form our knowledge of the architecture of the normal kidney.

#### TECHNIC

The details of the technical procedure require but a brief description. Pieces of a kidney that had been fixed and preserved in a dilute

This work was done with the support of the Josiah Macy Jr. Foundation.

From the Department of Pathology, Hoagland Laboratory, Long Island College of Medicine.

1. Oliver, J., and Lund, E.: Arch. Path. 15:755, 1933.

2. Huber, G. C.: Am. J. Anat. (supp.) 4:1, 1904.

3. Peter, K.: Untersuchungen über Bau und Entwicklung der Niere, Jena, Gustav Fischer, 1927.

solution of formaldehyde U. S. P. (1:10) or in Kaiserling's solution<sup>4</sup> were placed in concentrated hydrochloric acid and allowed to stand at room temperature until sufficiently softened. The time required depends on various factors, such as the degree of fibrosis that exists in the tissue, the size of the block under treatment, the length of its fixation, which may be years, and the temperature of the room. In some cases maceration will be sufficient in two days, in others a week is required, but these variations are of little moment, since the process may be watched and stopped at the desired point. We found that fresh tissue is of no definite advantage, though it macerates more rapidly, and since we wished to examine again kidneys that we had been studying for many years by other methods only fixed material was available.

When sufficiently macerated the almost diffluent tissue is washed in repeated changes of distilled water and may be kept for several days. The final procedure is the separation of the individual units from each other under the binocular microscope by dissection with needles. Illumination was obtained by reflected light.

No attempt was made, except in one special case to be mentioned later, to mount and preserve the isolated structures. The slightest pressure, even that developing from surface tension if the object is removed to a shallow drop on a slide, distorts the contours of the delicate tubules. They were therefore studied suspended in the water in which they had been dissected. Camera lucida drawings were made for record and illustration, but as an integral part of the method stereoscopic photomicrographs were taken at a standard magnification of 20. Such stereograms are in many ways preferable for study to the original preparations. Practically no detail is lost in the recording of the unstained structures, and the contours and twists and turns of a distorted tubule are more readily followed when the specimen is fixed on the photographic plate than when it floats suspended in water.

#### MATERIAL

The kidneys examined were all from cases of terminal hemorrhagic Bright's disease (chronic glomerular nephritis) in a series whose clinical and pathologic features have been previously described.<sup>5</sup> In the present study we are not interested in the topographic morphology of the individual nephritic kidney, but rather in a description of the various abnormal forms which the nephron may assume as a result of the chronic processes concerned. In a later study the combinations that are found in typical examples of the disease will be considered. The details of

4. This solution consists of 30 Gm. of potassium acetate, 10 Gm. of potassium nitrate, 750 cc. of distilled water and 300 cc. of a solution of formaldehyde.

5. Addis, T., and Oliver, J.: The Renal Lesion in Bright's Disease. New York, Paul B. Hoeber, Inc., 1931.

clinical history and of the general histologic characteristics of the kidney as seen in ordinary sections are therefore not given, but may be obtained if desired for the particular structures that have been illustrated by reference to the descriptions of the plate figures.

For simplification and completeness of description the course of the nephron from the glomerulus to the exit of the terminal ducts of Bellini into the pelvis will be followed, and the variations observed in each constituent portion will be described and compared with the normal.

#### THE NORMAL NEPHRON

Since a relative change in size is an important feature of the pathologic alterations to be described, the divisions of the human nephron as isolated from the normal kidney of a young adult are illustrated for the purpose of subsequent comparison. In figure 2 an intact complete short loop nephron from the glomerulus to the collecting tubule is shown. For a detailed description of the characteristics of the various segments of the normal unit reference may be made to Peter's monograph.<sup>3</sup>

#### THE NEPHRON IN TERMINAL HEMORRHAGIC BRIGHT'S DISEASE

*The Glomerulus and Its Vessels.*—The vessels of the glomeruli of the normal kidney are exceedingly delicate so that only with great difficulty can they be preserved intact with the isolated glomerulus. Their walls are thin and translucent and their contours even and tapering (fig. 1). In the abnormal kidney<sup>6</sup> they may be tremendously thickened, a fact evidenced by the comparative ease with which one obtains sprig-like portions of the terminal artery on which the diseased glomeruli hang in abundance by means of their afferent arterioles (figs. 3 to 7 and 13). The artery appears opaque with fusiform dilatations alternating with angular constrictions (figs. 3, 4, 6 and 13), and areas of fatty change, dazzlingly white, are scattered at random along its course in the form of diffuse mottled patches (figs. 7, 13 and 22).

From such sclerosed arteries spring the afferent arterioles; they too are thickened and rigid and at times flecked with bright patches of fatty change. Irregularity of contour, even aneurysmal-like bulgings are seen (fig. 13). Springing from the arterioles at an obtuse or even a right angle is frequently found Ludwig's vessel, dilated to a caliber equal to that of the parent arteriole (figs. 3, 6, 22 and 28). The significance of the size of this shunt across the occluded capillary bed of the glomerulus and the frequency with which it can be demonstrated will be mentioned later in considering certain of the changes that occur in the tubular apparatus.

6. For the sake of brevity the term "abnormal kidney" is used to denote the kidney in terminal hemorrhagic Bright's disease.

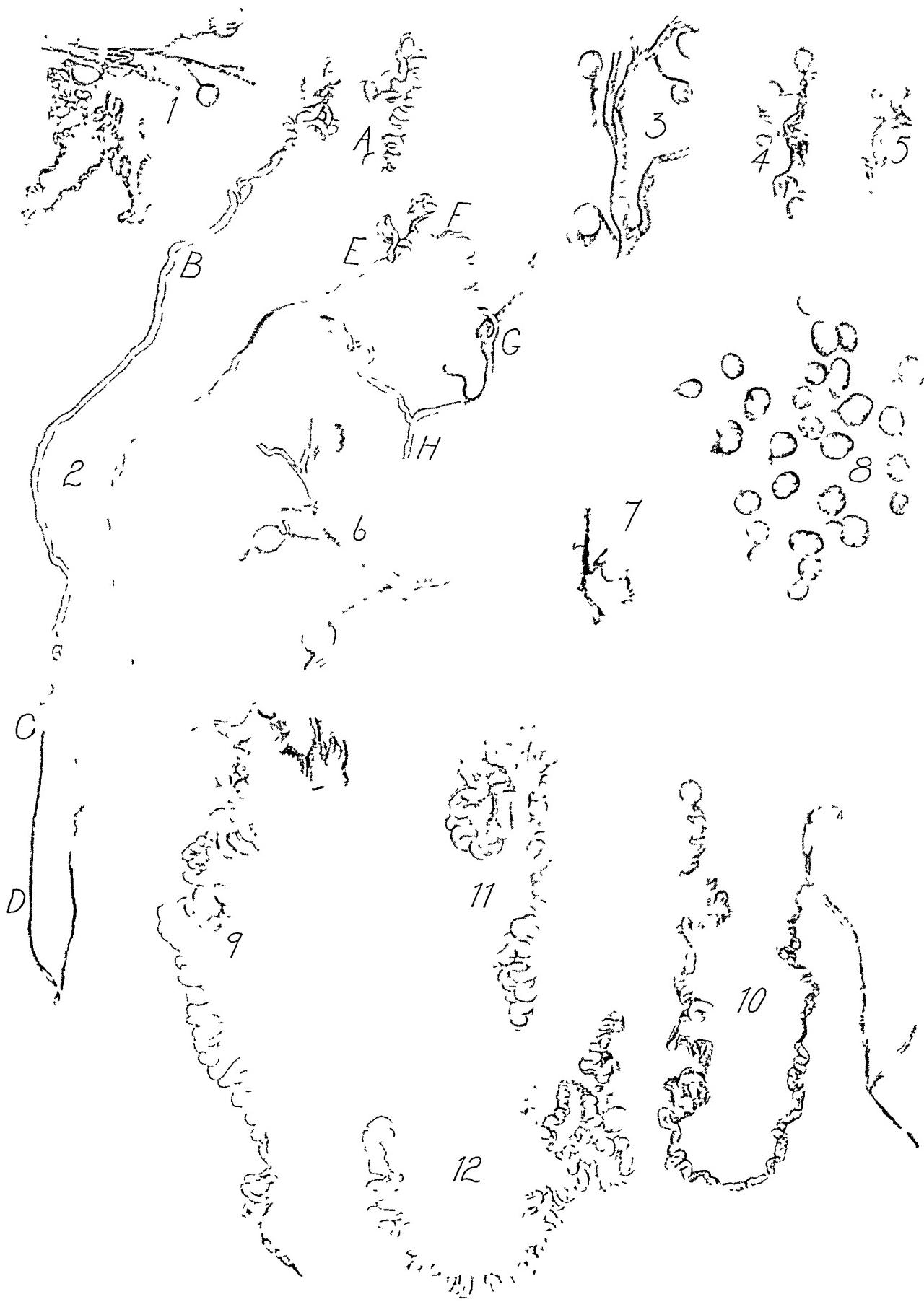


Fig. 1.—Terminal branches of an interlobular artery from a normal kidney showing glomeruli, to one of which is attached the first part of the proximal convolution. Another shows both an afferent and an efferent vessel;  $\times 15$ .

Fig. 2.—A complete nephron from a normal kidney. The junction of several other nephrons with the collecting tubule is shown. *A* to *B*, proximal convolution, periglomerular cluster; *B* to *C*, proximal convolution, terminal segment; *C* to *D*, narrow portion of Henle's loop; *D* to *E*, broad portion of Henle's loop; *E* to *F*, distal convolution; *F* to *G*, connecting piece; *G* to *H*, peripheral collecting tubule;  $\times 15$ .

Fig. 3.—Figures 3 to 61 depict kidneys with terminal hemorrhagic Bright's disease. Figure 3 shows terminal branches of an interlobular artery with glomeruli. Note the irregularity in size and the fat content of the tufts. Ludwig's vessel is apparent in the afferent vessel of the upper glomerulus on the left;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 36, p. 343).

Fig. 4.—A similar vessel showing atrophied and enlarged glomeruli;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 37, p. 348).

Fig. 5.—Irregularly dilated interlobular artery with fatty degeneration that extends into the afferent vessels;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 27, p. 302).

Fig. 6.—Dilated terminal artery showing glomeruli with Ludwig's vessel (middle glomerulus) and a long efferent vessel;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 36, p. 343).

Fig. 7.—Terminal artery showing rigid sclerosis and extreme fatty change. The glomerular tuft is also diffusely infiltrated with fat;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 15, p. 238).

Fig. 8.—Isolated glomeruli showing various forms of distortion and irregular fatty change in the tufts;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 36, p. 343).

Fig. 9.—A glomerulus greatly enlarged with a complete hypertrophied and hyperplastic terminal proximal convolution. The distal convolution, empty and dilated, is shown attached to the side of the glomerulus;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 6, p. 197).

Fig. 10.—Complete short loop nephron from glomerulus to collecting tubule. There is a marked hypertrophy and hyperplasia of the proximal convolution. The narrow segment of Henle's loop is a short straight segment;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 32, p. 325).

Fig. 11.—Markedly hypertrophied and hyperplastic proximal convolution. Note the kinked loops of its terminal portion;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 32, p. 325).

Fig. 12.—Another complete hypertrophied and hyperplastic proximal convolution with extreme kinking of terminal segment;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 32, p. 325).

The efferent vessel is much more difficult to demonstrate. This is partly due to the fact that even in the normal kidney it is not as definite and as well formed as the afferent vessel, for on its emergence it usually breaks up almost immediately into the capillaries of the intertubular network. It must be remembered too that in the great majority of the diseased glomeruli reduction or obliteration of the capillary bed in the tuft has caused a reduction in outflow and a consequent collapse of the efferent vessel. There is, therefore, an exaggeration of the normal disproportion in size of the two vessels of the glomeruli in the abnormal kidney. The difficulty in finding collapsed efferent vessels thus goes hand in hand with the relative ease with which dilated shunting Ludwig's vessels are found. In every case the efferent vessel if present is found to be of smaller diameter than the afferent (figs. 6, 26 and 28), and its delicate wall does not commonly show the extreme changes of sclerosis or degeneration that are seen in the latter.

The most striking change that affects the body of the glomerulus is variation in its size. All degrees of decrease in volume, apparently limited only by the smallest space that the fibrosed tuft may occupy when it is finally reduced to hyaline collagen, have been observed (figs. 4 and 5). It may be increased to sixty times its natural volume (fig. 31). In examination of the isolated intact glomerulus by the present method the cellular details of the abnormal process occurring in the substance of its tissues are invisible so that the cause of the enlargement remains obscure. Both inflammatory infiltration and hypertrophy of the tufts are possible factors, and although one cannot determine in a given isolated example which of these is present, it is evident from its shape and fatty content that changes other than a purely progressive hypertrophy are occurring. A supplementary study of histologic sections of the same kidney shows that practically all large glomeruli in terminal hemorrhagic Bright's disease are heavily infiltrated with inflammatory cells, and few, if any, show only the simpler hypertrophic change.

Alteration in shape of the glomerular body from the regularly round or oval contour of the normal structure is an equally striking abnormality. Egglike corpuscles with elongation through any diameter, pear-shaped bodies that may be fixed at either end to the afferent vessel, the production of flattened surfaces and of fissures and irregular lobulation result in almost every conceivable distortion (figs. 3 to 8, 13, 17, 20 and 21).

Though its cellular constituents are invisible, in many cases the abnormality of the tissues of the glomerular body may be recognized. The normal glomerulus is of an even gray opaque texture, and since the tuft fills the capsule almost completely very little internal structure can be made out. The great majority of glomeruli from the abnormal

kidney, however, contain a greater or less amount of fat which reflects the incident light with dazzling brilliancy. The appearance may be that of diffuse stippling with bright dots, in some instances so densely massed as to give the effect of a brilliant droplet of snow suspended, about to fall, on the twig of its rigid, fatty tinged crystalline afferent vessel (fig. 7). In others flecks or patches form an angular pattern which outlines the contour of the distorted tuft as it lies in its capsule; in still others the greater part of the body may be free with a single irregular broad patch of brilliancy (figs. 3, 5, 6, 8, 17, 27, 28 and 29).

As has been stated, the tuft itself is rarely seen in the normal glomerulus and then only with difficulty because it fills Bowman's capsule so completely. In the abnormal glomerulus, however, the capsule space is frequently distended and, as a result, the outline of the diseased tuft can easily be made out. All degrees of distention may be observed, from the production of a narrow irregular crescentic clear space, broadest at the tubular pole and decreasing toward the point of attachment of the tuft by its vessels, to conelike caps from the apex of which the tubule springs (fig. 6) or balloon-like appendages from which no tubule can be seen to arise (fig. 3).

The certain recognition of glomeruli whose tubules have been disconnected by the abnormal processes is difficult, for the attachment of the tube is normally weak, and in spite of considerable care fracture may occur during dissection. However, attention can be directed to such an atubular glomerulus before and during the dissection and the original absence of an attached tubule noted. When freed from the surrounding tissue the glomerulus may be turned with the needle and its entire surface inspected without the discovery of any interruption in the capsule. A careful examination is necessary in each instance before one can be certain that one is not dealing with an artefact. Even with this rigid examination many corpuscles were found that possessed no attached tubule. Further discussion of this observation will be given later.

#### THE PROXIMAL CONVOLUTION

Even in the normal kidney the general shape of the clustered mass of the proximal convolution is not constant. In the abnormal kidney, therefore, no characteristic change can be described for its general configuration. It is either smaller or larger depending on the nature of the change that has occurred in its constituent tubule. If there is a marked reduction in the diameter of the tubule there is usually a corresponding increase in the intertubular connective tissue, but such observations can be more clearly made in the reconstructed model<sup>1</sup> than in the teased specimen, for in the latter the tubule cluster is always more or less loosened from its original tightly packed condition. In

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Fig. 13.—The interlobular artery with afferent vessels to two glomeruli. From one arises the hypertrophied and hyperplastic proximal convolution. Its terminal segment shows a marked fatty change which does not, however, extend into the narrow limb of Henle's loop;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 25, p. 294).

Fig. 14.—The terminal portions of two hypertrophied and hyperplastic convolutions in which there is also dilatation. The kinks of the tubules have been drawn apart to show the degree of hyperplasia;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 32, p 325).

Fig. 15.—A complete hypertrophied and hyperplastic proximal convolution with fatty change in an isolated cluster of coils;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 25, p. 294).

Fig. 16.—A complete proximal convolution showing marked hypertrophy and hyperplasia;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 32, p. 325).

Fig. 17.—Glomerulus and portion of a proximal convolution, greatly dilated, showing a marked fatty change;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 36, p. 343).

Fig. 18.—Terminal portion of a proximal convolution that shows an extreme hyperplastic kinking and dilatation. There is also a marked granular degeneration of its epithelial cells. The first portion of the narrow limb of Henle's loop is considerably dilated;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 24, p. 288).

Fig. 19.—The terminal part of a hypertrophied and hyperplastic proximal convolution showing the complete narrow portion of Henle's loop and its passage into the broad ascending limb. The narrow segment of the loop is considerably dilated;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 37, p. 348).

Fig. 20.—Glomerulus with a proximal convolution which shows almost pure dilatation;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 25, p. 294).

Fig. 21.—Glomerulus with an atrophied proximal convolution. There is extreme irregular deformity of the tubule with marked fatty degeneration of its cells;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 26, p. 298).

Fig. 22.—Glomerulus and vessels with a complete proximal convoluted tubule that shows a marked hypertrophy. A very long Ludwig's vessel is shown;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 25, p. 294).

fact, the attempt was made in most cases to unravel the tubule purposely so as to show more clearly the details of its convolutions.

In the abnormal kidney the tubule presents little evidence of a "neck," the only exception being the production of a false neck in the examples in which the proximal convolution is greatly dilated with no corresponding thickening of its wall. The tubule contiguous to the glomerulus may remain undilated, thus forming a long narrow stretch that extends to the dilated portion (fig. 17).

The most striking changes noted in the proximal convolution are those of variation in size and fat content.

A reduction in diameter, the result of atrophy, may decrease the tubule to one half its normal caliber (figs. 21, 29 and 31). It is almost impossible to tease out these tenuous structures, especially since a fatty degeneration almost constantly accompanies the atrophy. Only fragments can be isolated completely. The best method therefore of studying these tubules consists in removing only partially the tissue which surrounds them. This is composed mainly of fibrous collagen which has softened and become translucent in the acid, and by the brilliancy of the excessive fat that is present in the atrophied epithelial cells the course of the shrunken tubule can be clearly observed and accurately followed. The loops are separated and widely spaced by the intervening scar tissue. This produces a simplification of the original complexity of the convolutions of the tubular mass and leads to such extremes as are seen in figures 29 and 31 in which the course of the tubules is almost straight.

The increase in the diameter of the proximal convolution may be due to actual hypertrophy with definite thickening of the wall of the tubule, to dilatation and a consequent thinning of the wall or to a combination of the two. If the enlargement is the result of hypertrophy, either alone or in combination with dilatation, there usually is an accompanying increase in the length of the tubule.

In the hypertrophied convolution an increase in diameter up to two and one-half times is frequently noted. The broad tubule preserves in a general way the normal arrangement of its convolutions (fig. 22). Throughout its length the tube appears either dense and solid, or with a narrow dark central streak which represents the lumen. Associated with this increase in the thickness of the epithelial cells there occurs in the majority of cases an increase in the length of the tubule. This is evidenced by the tortuosity of its convolutions, so that tightly formed secondary loops and kinks develop in the tubular mass which lies about the glomerulus. The cause of this lengthening of the tubule is the hyperplastic proliferation of its cells described in our previous study.

The complexity of the hyperplastic proximal convolution in the glomerular region is only an exaggeration of the normal condition, but

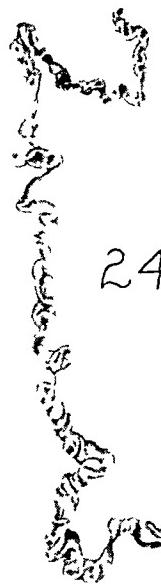
when the same process is noted in its terminal segment, which normally is a straight or at best a slightly undulating filament, a most striking appearance is produced. The thickened tube, at times twice its normal diameter, is folded back and forth on itself in tightly kinked, skeinlike bends, each so pressed on its neighbor as to have the appearance of a solidly fused irregular tube from which knoblike diverticula project (figs. 9, 11, 12 and 16). It is entirely impossible to follow the course of the tubule through these tortuous kinks, but they may be somewhat straightened out by stretching the tubule with the dissecting needles, and it is then seen that, although there are, in a certain sense, irregular projections from the walls, the tubule is in fact a continuous structure whose apparent diverticula are mostly due to its angular tortuosity. In figure 14 two enormous terminal portions of the proximal convolution are shown; these have been drawn out sufficiently to reduce their complex appearance. The passage into the narrow limb of Henle's loop may be seen at their lower ends.

The actual extent of the increase in the length of the proximal convolution by the development of these kinks is difficult to estimate. In typical examples of the normal tubule the terminal portion forms about one-third the length of the entire segment. In the abnormal kidney it is not unusual to find the hyperplastic tortuous terminal portion of equal length to the periglomerular mass. The significance of this disproportion will be considered later.

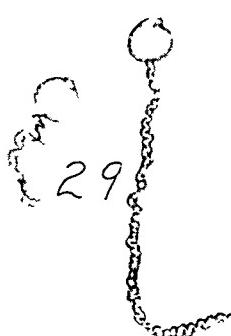
In the tubules just described there is little or no increase in the width of the lumen. In fact, the epithelial lining of the walls of many of them is so thick that the lumen cannot be seen at all. In others it is definitely dilated, at times measuring twice the normal diameter, but the wall is nevertheless thicker than normal. In these tubules hypertrophy and dilatation are evidently combined, and the result is the production of relatively enormous structures. Since hyperplastic lengthening is nearly always found accompanying the increase in thickness of the wall, such huge structures as that illustrated in figure 18 are not uncommon in certain kidneys. It will be noted that only the end fragment of a terminal portion of the proximal convolution is shown, and its passage into the narrow portion of Henle's loop is apparent. The periglomerular mass is, as a rule, relatively less massive, for it may be affected by the hypertrophy without marked dilatation.

Other proximal convolutions show pure dilatation of their lumens. The wall is therefore stretched to a membrane of almost endothelial-like thinness, which, when the tubule is isolated, falls in folds on itself as it collapses. It is extremely difficult to tease out these delicate structures in any great length, but figure 17 shows a part of an extreme example while figures 20, 26 and 28 present tubules showing varying degrees of dilatation without any thickening of their walls. Figures 23

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23



31



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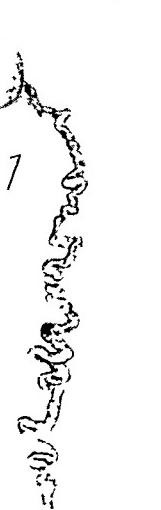


Fig. 23.—Complete short loop nephron. The glomerulus is greatly enlarged and the proximal convolution dilated with little evidence of hyperplastic kinking. The narrow limb of Henle's loop is dilated and passes into the broad ascending limb which ends in a dilated and empty distal convolution;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 27, p. 302).

Fig. 24.—A loop of Henle arising from a hypertrophied and hyperplastic proximal convolution;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 24, p. 288).

Fig. 25.—A loop of Henle from the long loop type of nephron, including the terminal portion of the proximal convolution. The narrow portion is moderately dilated. The ascending broad portion is irregularly dilated and contains some solid material;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 24, p. 288).

Fig. 26.—Greatly enlarged glomerulus with fatty change. The afferent and efferent vessels are well shown. The proximal convolution shows a simple dilatation. There is a marked fatty deposition in an isolated group of coils;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 36, p. 343).

Fig. 27.—A glomerulus with complete proximal convolution showing a combination of atrophy, dilatation, hypertrophy and hyperplasia;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 6, p. 197).

Fig. 28.—The glomerulus is enlarged. The efferent and afferent vessels are well shown along with Ludwig's vessel. The tubule is dilated and shows considerable fatty change;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 36, p. 343).

Fig. 29.—Two glomeruli with portions of markedly atrophied proximal convolutions. Note the simplification of the convolution pattern;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 23, p. 283).

Fig. 30.—Complete short loop nephron showing dilatation of the proximal convolution. The dilatation extends into the narrow limb of Henle's loop and its ascending portion and the convolution. The latter is empty;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 27, p. 302).

Fig. 31.—Tremendously enlarged glomerulus, 60 times the average normal volume. There is a moderate atrophy of the proximal convolution with simplification of the normal looping of the convolutions;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 38, p. 354).

Fig. 32.—The greater part of a proximal convolution showing hypertrophy, hyperplasia and dilatation combined. There is a marked fatty change in the terminal portion which ends abruptly as the tubule becomes the narrow segment of Henle's loop;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 14, p. 288).

and 30 are of interest since complete short loop nephrons from the glomerulus to the collecting tubule are intact. It is significant that such dilated broad tubules seldom show the hyperplastic kinking so common with the hypertrophic increase in diameter, but are characterized by a definite decrease in the complexity of their convolutions. In figure 20, though the original course of the tubule has been disentangled, it is readily apparent that no condition approaching the tortuosity shown in figures 9, 11, 12 and 16 could ever have existed.

Associated with dilatation, particularly in tubules which show this passive increase in diameter with no evidence of progressive hypertrophic change, fatty accumulations are seen in the thin epithelial walls (figs. 17, 20, 23, 26 and 30). The droplets of fat are usually diffusely sprinkled and may extend throughout the entire length of the isolated segment. In other instances the fat is sharply limited to a few contiguous coils (fig. 26), so that a loop passes out of the area of change, becomes free and then, reentering it, becomes again involved. Thus irregular small stretches of fatty deposit in the tubule, interrupted by normal segments of epithelium, are produced.

It is much less common to find fatty accumulations in the proximal convolutions that show increase in size from an actual hypertrophy of their walls. However, examples may be found, either with a restricted cluster of loops filled with refractile droplets (fig. 15) or with irregular small patches scattered along the greater part of the course of the tubule. Not infrequently it is the end part of the terminal segment of the convolution that is involved, the fatty metamorphosis beginning abruptly and extending downward to end with equal suddenness where the proximal convolution passes into the narrow limb of Henle's loop (figs. 13 and 32).

The change in the proximal convolution, whether taking the form of an atrophy, a hypertrophy, a dilatation or a hyperplasia, has been described so far as affecting this segment in a single manner, the result being the production of a tubule that is either greater or smaller than the original structure. This often occurs, but in many instances single units show a combination of all these changes. In figure 27 a proximal convolution is atrophic in its first part; it then assumes a normal caliber, which is increased by a definite hypertrophy, while its terminal portion is markedly kinked by hyperplastic growth. Similarly in figure 17 the first portion of the convolution is reduced in thickness but suddenly increases in diameter with a tremendous dilatation. All conceivable combinations of these changes have been observed and, along with the isolated occurrence of fatty change in limited and contiguous loops of the tubule, give evidence of the importance of local processes the significance of which will be considered later.

## THE LOOP OF HENLE

The division of the nephron into two types, those with long loops of Henle and those with short, can be easily made in the abnormal kidney. Although, as their names indicate, the length of the two types is a distinguishing feature, this characteristic is only relative, and the criterion for their identification in the human kidney is the location where the bend of the loop and consequent reversal of direction occurs. In the case of the long loop the bend is situated in the inner zone of the medulla. Since only the narrow segment of the loop reaches this depth, the bend is invariably in that part of the tubule. Short loops reverse their direction in the outer zone and even at times in the cortex. In these regions of the kidney the broad segment is present, and the actual bend always occurs in it. Examples of short-looped nephrons have been demonstrated completely; that is, the nephron from the glomerulus through the first part of the collecting system has been isolated intact by dissection (figs. 10, 23 and 30). It is important to note that in the human kidney the short loops are much more frequent than the long; according to the counts of Peter they are seven times more common. As has been the experience of those who worked with the normal human kidney, we have been unable to isolate a long loop completely, since the tenuous narrow portion is a structure of great length and fragility. All the various parts of these loops are, however, easily obtained.

## THE NARROW SEGMENT OF THE LOOP

The narrow segment which in the long type of nephron forms a portion of both the descending and the ascending limb retains its normal characteristics to a remarkable degree in the abnormal kidney. In short loops it may exist as a short clear segment of its usual diameter separating the hyperplastic proximal convolution from the broad limb of the loop (fig. 10), or in others it may, as happens in normal kidneys, be entirely missing.

A curious effect on the direction of the narrow segment is produced by hyperplastic processes in the terminal portion of the proximal convolution. Not all of the increase in length of the tubule is taken up by the kinking that occurs in it, and as a result there is a definite extension toward the medulla of the kidney. Peter has emphasized the fact that the actual bend of the loop in the short variety of the human nephron is always in the broad segment, and the normal direction of the narrow segment is, therefore, always descending. But in some hyperplastic short loop units there results an inversion of this direction of the narrow segment, for the advancing terminal portion of the proximal convolution grows down past the original bend of the loop, drawing the narrow segment after it. It is only by such a process that the appearance shown in figure 19 can be explained. In the latter the

whole length of the narrow segment is shown to its termination in the broad segment. The bend of the loop is situated in it, close to the end of the hyperplastic terminal portion of the proximal convolution, and its entire course is ascending, so that it forms the greater part of the distal limb of Henle's loop.

Narrow segments that lie closely pressed among the hyperplastic kinked terminal portions of the proximal convoluted tubule are often molded into a similar wavy course (figs. 19 and 24). Part of this kinking may be due to hyperplastic growth in the narrow segment itself, but the appearance noted during the dissection indicates that the greater part of the change is a passive one.

Peter describes the occurrence of crystals in the narrow segments of the loop. We have noted them—in some cases they are a definite brick red—but we have not noticed any significant increase in their amount.

#### THE BROAD SEGMENT OF THE LOOP

The actual transition from the narrow to the broad segment in the normal kidney is determined more by the character of the epithelial wall than by the caliber of the tube. The clear cells of the narrow portion are replaced by the darker opaque ones of the broad segment. But in the abnormal kidney in which degenerative changes are apt to occur throughout the epithelial lining of the tube the exact point at which a narrow segment becomes broad may be indistinct. In figure 10, depicting a complete short loop nephron with hypertrophy of the proximal convolution, the relatively clear appearance of the narrow segment is preserved so that the division is quite apparent. In figure 23 another complete nephron is illustrated. From the end of the dilated fatty proximal convolution arises the narrow segment which passes downward to bend abruptly back forming the loop. From there the ascending distal limb, comprising the narrow and broad segment, follows, but it is difficult to determine the exact point of transition between the two. The broad limb finally ends in the distal convolution. It is somewhat tortuous, but the contours of its walls are even. In figure 25 a narrow segment from a long loop containing the bend is shown. It grows increasingly thicker as it ascends to change without any exact line of demarcation into the broad segment. The latter is only slightly increased in caliber, and save for the moderate tortuosities is essentially normal.

Tortuosities or varices and dilatation are the most frequent abnormalities in the broad segment. We have never found any appearance that could be interpreted as evidence of hypertrophy or hyperplasia. In figures 24, 25, 39 and 42 varying degrees of varix formation are present in segments otherwise only moderately dilated. The different appearance of these irregularities from the even hyperplastic kinking observed

in the proximal convolution is obvious. Not infrequently the irregular dilatations contain coagulated material, which increases in amount until the distal convolution is reached. This portion of the nephron, as we shall see later, is commonly filled completely with solid substance.

A more even and general dilatation of the broad segment with a consequent thinning of its wall is also frequently observed. In figure 30 a short loop nephron shows the proximal convolution ending in an extremely short narrow segment. The bend of the loop occurs in the broad segment. This portion, greatly and fairly uniformly dilated, is apparently empty<sup>7</sup> and ascends in broad waves to pass into the distal convolution.

Whether or not dilatation occurs behind an obstruction of the tubule appears to depend on more factors than are included in the obstruction to flow alone and will be discussed later. It will be sufficient at present to note its varying occurrence in the more distal portion of the tubule in which plugging of the lumen becomes progressively frequent. In figures 33, 38, and 43 varying degrees of dilatation of the ascending limb are present, and in most examples the distal convolution contains solid material. In the broad segments that show the greatest degree of dilatation, however, the widened lumen is not empty but is filled with a translucent coagulum so transparent that at first glance the tubule appears empty. In other instances the clear substance is flecked with minute striations of more opaque material that gives a feathery appearance to the whole mass. At times (fig. 44) the distention is tremendous, the diameter of the segment being four times the normal. The wall is greatly thinned, and the smooth contours of the tube swell and project in a manner entirely different from the irregular varices previously described.

To sum up the abnormalities found in the loop of Henle, including both its narrow and broad segments, it may be stated that only passive changes have been noted, especially distortion and dilatation. No hyperplastic or hypertrophic changes were observed, the only exception to this statement being the secondary, and therefore in a sense passive, lengthening of the narrow segment when it is drawn downward by the actively growing hyperplastic terminal portion of the proximal convolution. There is a great contrast, therefore, between the pathologic processes concerned in these two different divisions of the nephron.

#### THE DISTAL CONVOLUTION

The term "distal convolution" is in one sense unfortunate for it gives the impression that this portion of the nephron is the morphologic

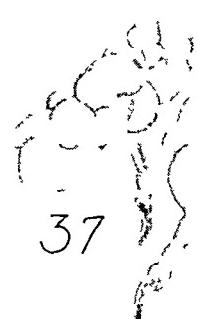
7. The term "empty" is applied to the appearance of lumens that were formerly filled with clear fluid or urine that has left no trace in the fixed material.



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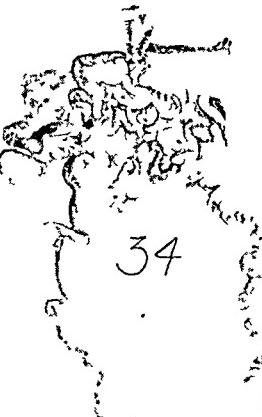
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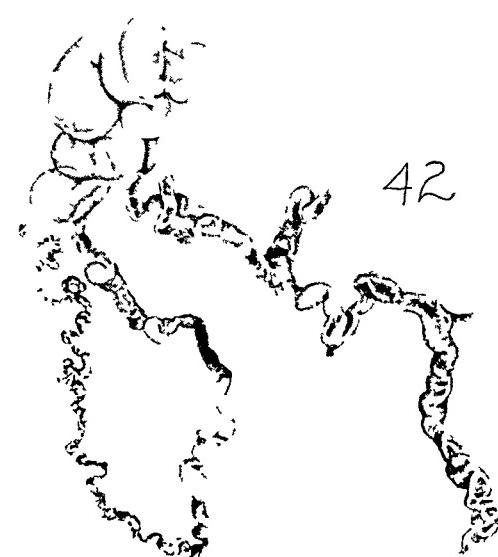
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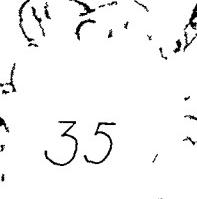
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Fig. 33.—Tremendously enlarged distal convolution packed with heterogeneous débris. The ascending limb of Henle's loop is greatly dilated and empty. The dilatation extends around the bend into the narrow limb of the loop;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 25, p. 294).

Fig. 34.—The glomerulus and the periglomerular mass of the proximal convolution. There is a considerable fatty degeneration in this portion of the tubule. At the left the broad limb of Henle's loop ascends to the region of the glomerulus where a dilated empty distal convolution finally passes to the right by its connecting tubule;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 42, p. 370).

Fig. 35.—At the left a dilated ascending limb of Henle's loop passes into the distal convolution, followed by the connecting piece and collecting tubule. All these tubules are packed full of highly refractile granular débris. The lower part of the collecting tubule is, however, constricted and empty;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 37, p. 348).

Fig. 36.—Greatly enlarged glomerulus to which is adherent a distal convolution that is in part empty but a few loops of which contain granular débris;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 27, p. 302).

Fig. 37.—On the right the ascending limb, tremendously dilated with refractile material, passes into a dilated distal convolution stuffed with the same type of débris. The connecting piece runs directly down from the cluster of its coils;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 27, p. 302).

Fig. 38.—The distal convolution above is packed with débris. The ascending limb in Henle's loop is dilated and empty, as is the connecting piece;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 27, p. 302).

Fig. 39.—Another distal convolution irregularly filled with material. Note the adherent glomerulus;  $\times 15$  (Addis and Oliver,<sup>5</sup> case, 27, p. 302).

Fig. 40.—On the left the entrance to a distorted plugged distal convolution that ends in the connecting piece on the right;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 36, p. 343).

Fig. 41.—A typical deformity in the plugged distal convolution. On the right is the empty connecting piece;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 36, p. 343).

Fig. 42.—A short nephron complete except for the glomerulus and portion of the proximal convolution. The latter shows a marked atrophy and fatty degeneration. The ascending limb of Henle's loop passes into a greatly distended distal convolution which is packed and distended with débris. The connecting piece and place of origin of the collecting tubule are empty;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 27, p. 302).

equivalent of the proximal convolution. As Peter showed, this is far from being the case, since the distal convolution consists at best of a few poorly formed loops, never approaching in complexity the complicated system of the proximal convolution (fig. 2). The term is, however, firmly established in English nomenclature, and we shall therefore use it, although it will be apparent in the presentation that follows that just as in the normal kidney the two convolutions, proximal and distal, are morphologically different, so in the abnormal organ they differ strikingly both as to the nature of the processes which affect them and in the final structural result produced.

The broad limb of Henle's loop passes into the first irregularities of the distal convolution at the level of the glomerulus from which the nephron arose. As Peter has shown, the tubule is attached quite firmly at this point to the capsule of the corpuscle and is frequently found still adherent to it when isolated by dissection. The same is true in the abnormal kidney (figs. 9, 36 and 39).

The most common abnormality in the distal convolution is simple dilatation. Gradually increasing distention distorts the lumen until its diameter may be over five times that of the normal (figs. 9, 23, 30, 34 and 45). The wall, greatly stretched, shows no sign of a compensatory thickening but is almost transparent. Figure 34 shows such a dilated distal convolution *in situ* with its relations to the glomerulus and the proximal convolution. The latter is filled with fat, and thus contrasts strongly with the translucent thin-walled distal segment with which it is entangled.

In the normal kidneys studied, which are all from young adults, we have found very few of the varicose distal convolutions described by Peter, while in the abnormal kidneys irregularity is consistent and exaggerated far beyond anything that he observed. It is interesting to note in passing, therefore, that he found such structures with increasing frequency in the kidneys of older persons. In the abnormal tubule the irregularities take the form of fusiform swellings that taper to narrow necks, bulbous varices that jut out in grape cluster fashion from the tubule, or spherical appendages (figs. 9, 23, 30 and 41).

The other abnormality noted in the distal convolution is found intimately associated with dilatation. It consists of a filling of the distended and distorted loops with solid débris so that the flexible tubule which normally bends and sways with the motion of the slightest current in the suspending water is transformed into a solid inflexible tortuous structure. The least attempt to rearrange its convolutions with the dissecting needles results in the fracture of its brittle substance. The solid material consists of a varying mixture of fatty substances that are highly refractile and irregular granular masses which have the

appearance of coagulated albuminous material. All degrees of occlusion occur, from a stuffing of a single loop or a diverticular appendage of the main tube to solid packing of complete convolutions (figs. 37, 38, 39, 42, 43 and 46). Examples are also found in which alternation of empty and plugged loops occurs (fig. 40). The filling of the lumen may extend beyond the limits of the distal convolution downward into the broad ascending limb of Henle's loop, an appearance that has been previously described (figs. 35 and 39).

A characteristic of the solid material that fills the lumen of this portion of the nephron is its lack of homogeneity. Its density varies irregularly, more solid opaque masses alternating with lighter substances. Adjacent loops, for example, often show this appearance strikingly (figs. 39, 40 and 43). Another peculiarity is the sudden and abrupt ending of these deposits. A sharp end of solid substance, either straight or rounded, projects into the dilated portion that is free from débris. This is particularly well illustrated in figure 33, which shows a veritably gargantuan distal convolution packed full of heterogeneous material. At both limits of the accumulation the sudden cut-off ends abut into the empty portion of the tubule. The usual variation in the density of the occluding substance is also evidenced in this example.

The common accompaniment of the collection of débris in the distal convolution, namely, the dilatation of the fluid-containing, broad limb of Henle's loop, is apparent in figure 33. It is stretched by internal distension to at least four times its normal diameter, and even the narrow segment of the loop proximal to the bend is definitely distended.

Solid material of an entirely different character from this heterogeneous mixture of fat and granular substance is also found in the dilated distal convolutions. This consists of the almost perfectly translucent jelly-like material that has been described as at times filling the distended broad limb of Henle's loop. It may, in fact, fill a considerable length of tubule, including the dilated broad limb of Henle's loop, the distal convolution, the connecting tubule and a large portion of the collecting tubule (fig. 44), yet throughout show no variation in its clear translucency. Moreover, there is never any interruption of it from loop to loop as was not infrequent in the distal convolutions that were filled with the heterogeneous débris-like material. As will be emphasized later, it appears as if the contents of a tubule dilated and filled with a coagulable fluid had congealed at a given definite time throughout its entire extent.

Peter describes the occurrence of large numbers of crystals in the distal convolution. We have seen them, but have found no excessive collection of them. It may be that in our abnormal material they are obscured by inclusion within the dense opaque débris.



Fig. 43.—A plugged distal convolution with marked dilatation and only partial filling of the ascending limb below. Above, a dilated connecting piece and collecting tubule;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 25, p. 294).

Fig. 44.—Tremendously dilated ascending limb, distal convolution, connecting piece and collecting tubule. The entire tubule is distended with clear coagulated jelly-like material;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 35, p. 339).

Fig. 45.—Two distal convolutions and their connecting tubules joining to form the collecting tubule. Both are greatly dilated but empty;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 27, p. 302).

Fig. 46.—Three distal convolutions, two plugged and one empty, and their junction to form a collecting tubule;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 27, p. 302).

Fig. 47.—Traplike loops in the peripheral collecting tubules. The loops are filled with refractile débris;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 37, p. 348).

Fig. 48.—A peripheral collecting tubule containing a castlike mass of solidified débris. Note the constriction below the occluding material;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 32, p. 325).

Fig. 49.—Irregularly dilated and constricted peripheral collecting tubule. One branch is filled with castlike substance;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 37, p. 348).

Fig. 50.—Undeformed peripheral collecting tubules containing a hyaline cast. Contrast the ease with which such a cast might escape with the conditions of the previous three specimens;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 42, p. 370).

Fig. 51.—Sacculated deformity of peripheral collecting tubule;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 32, p. 325).

## THE CONNECTING TUBULE

Peter describes the *Schaltstück*, or distal convolution, as consisting of two divisions, the first and the second. The latter, differing very little from the first in its structure, passes into the origin of the collecting tubule. This second division may be called the connecting piece, for there is a certain advantage in preserving its individuality in describing the nephron of the abnormal kidney. In most cases it consists only of the less twisted portion of the tubule that joins the definite loops of the distal convolution to the collecting tubule, which also does not differ greatly from it. The separation into these several parts is made chiefly by the general configuration and the direction of a continuous tubule. This is particularly true in the abnormal kidney when all these portions are dilated or when they are filled with what appears as a clear coagulum (fig. 44).

In many instances, however, the connecting piece is not dilated, and therefore stands out in marked contrast to the greatly dilated distal convolution that precedes it. Especially is this contrast striking when the loops of the convolution are filled with mixed débris and this material ends abruptly to leave the constricted connecting piece empty (figs. 33, 38, 40, 42 and 43). In such cases the large distorted and plugged coils of the distal convolution end at both extremities in narrow, empty tubes, proximally in the broad limb of Henle's loop and distally in the connecting piece.

## THE COLLECTING SYSTEM

The origin of the collecting system is readily observed in the abnormal kidney, perhaps even more easily than in the normal organ, for the dilated distal convolutions stand out prominently when filled with solid material, and their passage by means of the connecting tubule into the peripheral collecting tubule is plainly evident. In figure 45 an enormously dilated but empty distal convolution joins by means of its connecting piece with another connecting piece the convolution of which is broken away and so forms the collecting tubule. In figure 46 the connecting piece of two distal convolutions join to form the duct to which a third convolution is soon added.

The collecting tubules just described have appeared as empty tubes. Not infrequently those distal convolutions that are greatly distended with translucent jelly-like coagulum pass into a collecting tubule which is equally dilated and filled with a continuous mass of the same material (figs. 44 and 54). The tubule shown in the lower portion of figure 54 contains mixed débris.

The collecting tubule may also contain, from its origin, the heterogeneous fatty and granular substance that so frequently fills the distal convolutions. In figure 35 the ascending limb of Henle's loop, the distal convolution, the connecting tubule and a great length of the col-

lecting tubule are shown to be densely packed with débris. The whole structure when isolated was rigid and easily fractured. A connecting tubule from another nephron containing the same material joins the duct. Accumulated matter has irregularly distended its lumen, and this distention ends suddenly while the tubule continues diminished in caliber and empty. Such dilatations filled with solid material lying proximal to an empty lumen of lesser diameter are frequently found in the collecting system.

As in the normal kidney, the peripheral collecting tubules continue independently down through the depths of the cortex and in the outer zone of the medulla join to form the central tubules. From there on a repeated union of ducts by the typical tuning fork junction described by Peter lessens their number with an increase in the size of the lumen of the resulting tubule.

In its course through the cortex the peripheral collecting tubule may be joined by the connecting piece of an occasional distal convolution. Even in the normal kidney its course is, however, not always direct, for knotlike kinks occur as well as retrograde loops which turn back toward the cortex for a short distance and then return to their medullary direction. In the abnormal kidney these kinks and loops assume a considerable importance because they are often filled with solid granular material similar in appearance to that observed in the distal convolutions (fig. 47). The tubule in the loop is distorted and thrown into irregular bulging segments as if it had been actually stuffed by accumulating débris under a considerable pressure. One gains the impression that the loop has acted as a trap for material carried to it by the stream of the urine, for the lumen below the obstruction is smaller than that occupied by the occluding plug.

The normal central collecting ducts which first arise at the junction of the outer and inner zone of the medulla are evenly contoured tubes the lumens of which become larger with each fusion. Fairly normally preserved ducts of this character may be found in the abnormal kidney, and within them may be seen solid material firmly molded into clear hyaline casts of the lumen (fig. 50). But the majority of the central ducts of the abnormal kidney lack their normal characteristics, for they are irregular both in contour and in caliber. The irregularity of their contour consists of increases or decreases in diameter that alternate in no definite order. The lumen of such ducts may or may not contain débris (figs. 48 and 49).

A more striking appearance is offered by the ducts in which the dilatation and constriction affect the contour of the tube in a more regular and patterned fashion. Such distortions may take the form of sacculated bulgings, at times almost spherical, while in other instances they are blocklike, or all manner of irregular transitions from one type



Fig. 52.—Large central collecting tubule showing the typical fusiform sacculated deformity. The segments are filled with débris. Note the narrow constricted portion below;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 32, p. 325).

Fig. 53.—Central collecting tubule showing fusiform and spherical deformity with irregular constrictions of the lumen;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 32, p. 325).

Fig. 54.—The origin of a collecting tubule. The upper portion is dilated and filled with homogeneous, clear, jelly-like material. In the lower portion an obliteration by refractile débris fills the lumen;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 37, p. 302).

Fig. 55.—Irregularly deformed central collecting tubules filled with débris that contains crystals and a spherolith;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 32, p. 325).

Fig. 56.—Collecting tubules showing the fusiform type of deformity. One tubule is filled partially with débris and crystals; the others are empty;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 32, p. 325).

Fig. 57.—The termination of the collecting system in a duct of Bellini. Note the casts filling the terminal collecting tubules. The footlike process is composed of the epithelium which lines the operculum;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 6, p. 197).

Fig. 58.—Terminal portion of a dilated hypertrophied and hyperplastic proximal convolution that has been separated by the pathologic processes into three segments. The lower one continues on into the narrow portion of Henle's loop;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 6, p. 197).

Fig. 59.—Cystlike structures resulting from multiple disruption of tubules. They are filled with coagulated material;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 36, p. 343).

Fig. 60.—Terminal portions of the proximal convolution showing extreme deformity with sacculation;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 6, p. 197).

Fig. 61.—Isolated vesicles resulting from the disruption of hypertrophied and hyperplastic terminal portion of the proximal convolution;  $\times 15$  (Addis and Oliver,<sup>5</sup> case 6, p. 197).

to the other are observed in the course of a single tubule (figs. 51, 52, 53, 55 and 56). Such distorted ducts may be empty. In many examples the sacculation is so extreme that the duct is transformed into a segmented flattened structure three times as wide as it is thick. The segments are connected to each other by narrow communications and are arranged so that their size uniformly increases to a maximum perhaps four or five times that of the original duct and then decreases until the normal diameter of the tubule is again reached. In this manner a long fusiform sacculated dilatation of the tubule is formed which is nearly always filled to a greater or less extent with granular débris. In the débris crystals may be found in great number, some colorless minute needles, others a bright red. Spheroliths also are not uncommon (figs. 55 and 56).

The number of the central collecting ducts is eventually reduced by fusion, and they join to form the terminal duct of Bellini. Peter was unable to isolate this structure in the human kidney because of the fragility of its thin wall that encloses so great a cavity. In the abnormal kidney it is usually packed with heterogeneous débris so that its contents support its walls, and this facilitates dissection. In figure 57 the larger collecting ducts rapidly join by pairs until three large trunks are formed. These three trunks unite to form the duct of Bellini, a bulbous structure which constricts and then opens into the pore of the area cribrosa. A portion of the epithelial lining of the pore has remained attached to the margin of the opening of the duct.

The terminal collecting tubules contain solid material in masses which from their definitely molded cylindric form may be designated as "casts." The contours of these large tubules are not distorted, and it seems quite possible therefore that such casts might be swept with the urine into the bladder. The capacious cavity of the duct of Bellini is also filled, though its content is not consolidated into a definite plug or "cast" of the lumen, but consists of loosely packed clumps of granular débris.<sup>8</sup> Thus are formed the huge but fragile "renal failure casts" the appearance of which *in situ* has been previously illustrated in histologic section.<sup>8</sup>

And so, reaching the pelvis of the kidney, ends the course of the abnormal nephrons and their terminal ducts.

#### STRUCTURES PECULIAR TO THE ABNORMAL KIDNEY

The structures so far described, though in many instances greatly distorted from their original morphologic condition, retain the fundamental characteristics of the normal nephron. Other structures found in the abnormal kidney, though derived from normal architectural units

8. Addis and Oliver,<sup>5</sup> fig. 17, p. 139.

by the action of the pathologic processes, nevertheless are peculiar to it, for they have no counterpart in the normal organ. The production of these abnormalities is due mainly to the action of the chronic and progressive inflammatory process which, occurring in the interstitial frame-work of the organ, exerts a continuously increasing pressure as the products of the reaction increase. To this deleterious factor is added the disrupting and distorting force that develops when the inflammatory tissue begins to contract. The distortions of the nephron heretofore described were due in part to such forces; the most severe result of them is an actual disruption of the unit.

We have already described how the glomerulus may be pinched from its tubule and lie free as an isolated object still attached to its vessels. If the tubule may be interrupted at its exit from the glomerular body, a point of structural weakness, such an interruption is also possible at any place or at repeated places in its course.

In our earlier histologic study by means of sections of these same kidneys we found certain cases in which large areas of the renal parenchyma were composed of closely grouped cross-sections of tubules. These areas occurred most frequently in the outer zone of the medulla but were also present in the cortex. The tubules that filled them were lined by an intact epithelium of an atrophic and atypical appearance, and their relatively large lumens contained coagulated material that was hyaline, though central condensation and lamination of its substance could at times be seen. Figure 62 shows the appearance in section of these tubules, and another example may be seen in the previously published monograph.<sup>9</sup>

There is something about the arrangement in the section of these tubules that arouses the curiosity if not the suspicion of the observer as to their actual nature. When examined by microdissection the reason for their peculiarity is apparent (fig. 59). The "areas filled with tubules" are not groups of closely packed tubes but large masses or irregular collections of small round or oval cysts, the diameters of which vary within the limits observed in the tubular structures of the abnormal kidney. They lie in closely clustered masses of considerable size that may contain no other elements of the original renal structure. When macerated material is examined in which the action of the acid has only softened and made translucent the collagenous material of the scar tissue that always surrounds them they appear not unlike the jellied egg clusters of certain Amphibia. Each cyst contains solid clear material which, as in the stained histologic section, may show central condensations.

Any doubt that these cysts arose from the disruption of tubules is removed by a careful examination of their shape. Although the major-

9. Addis and Oliver,<sup>5</sup> fig. 38 A, p. 356.

ity are perfect spheres or oval bodies, examples that are definitely elongated fragments of tubes are found occasionally. Others of considerable length are irregularly fusiform or pear-shaped.

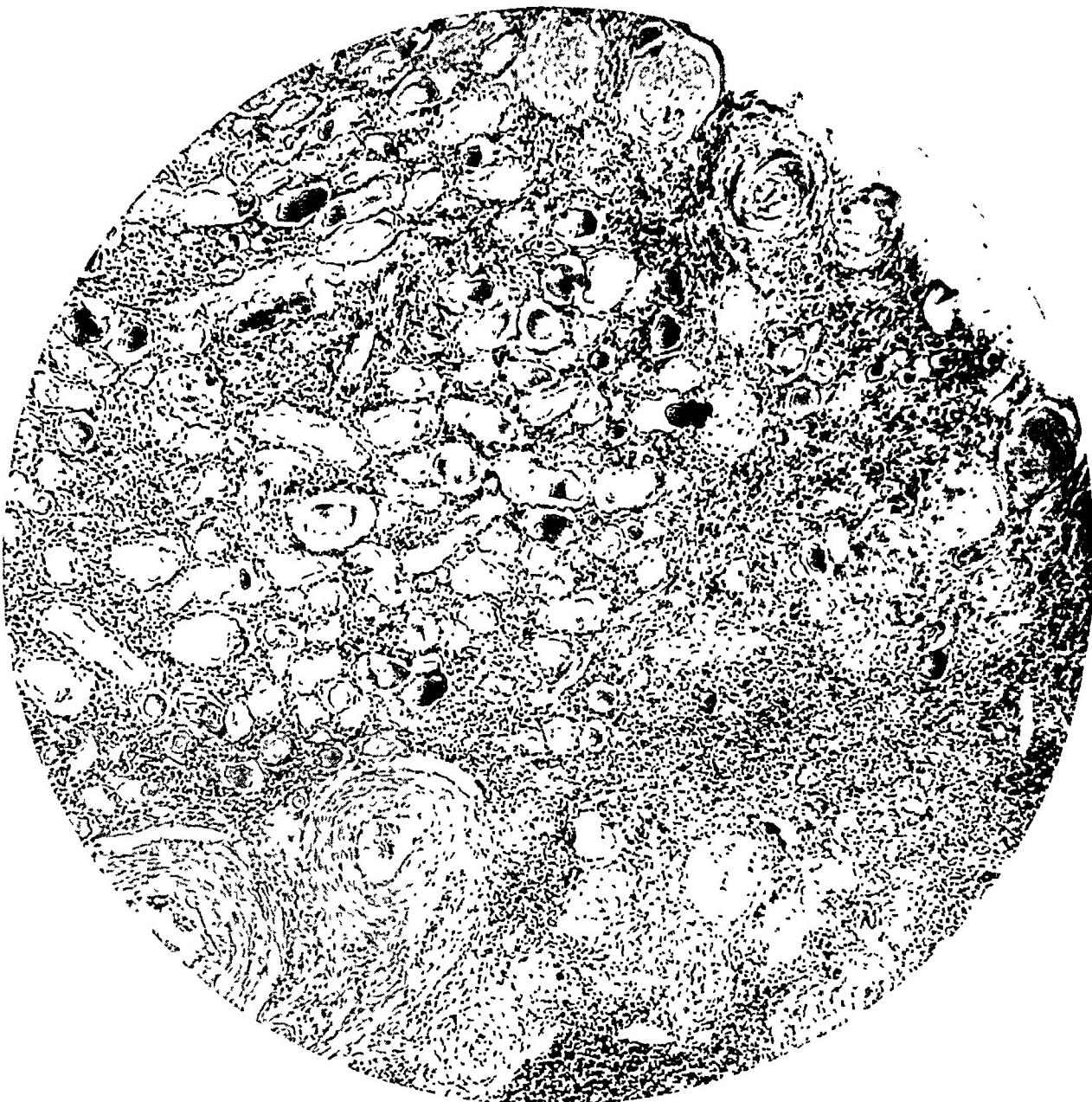


Fig. 62.—Section through the kidney from which the isolated cysts shown in figure 59 were obtained. The "tubular" appearance of these cysts in histologic section is shown at the upper left hand part of the field.

None of the kidneys studied showed cysts whose size was sufficient to make them evident by gross examination. Even in the histologic section it was apparent that most of the so-called "microscopic cysts" were only sections through greatly dilated but continuous tubules, and

our descriptions of isolated tubules, in particular of the distal convolutions, show how enormous such dilatations may be. The occurrence of true cysts of small diameter can be certainly determined, therefore, only by the actual isolation of the dilated structures.

It is difficult to decide exactly what portions of the tubule contribute to the formation of these cysts. They lie in regions of the cortex or medulla that contain all the various portions of the tubule. Their epithelial lining, examined in section, is so simplified by atrophy that it no longer presents any evidence for identification, but from the appearances seen in figure 62 and in figure 38A of our previous study<sup>9</sup> one can at least be certain that both convoluted and "straight" tubules have been their source.

Besides cysts which contain within their walls a definite cavity, similarly rounded structures are found scattered through the areas of tubular disruption that are even smaller in diameter but entirely solid. Observed beneath a cover slip by higher magnification they are found to be clusters of plump, epithelial-like cells that frequently contain fat. Their situation among the products of tubular destruction and their rounded contours leave no doubt that they are masses of tubular epithelium in which no lumen persists.

Another form of tubular disruption, but one limited to a specific portion of the tubule, was also not recognized in our former study of this material by histologic sections. When the deeper layers of the cortex and the outer stripe of the outer zone of the medulla are dissected, among the hypertrophied and hyperplastic terminal spiral portions of the proximal convolution are found isolated segments of tubule the external contours and general configuration of which resemble exactly those of the intact enlarged kinked spiral (fig. 61). They are obviously interrupted or pinched off fragments of this terminal portion of the proximal convolution (fig. 60), for they may be found lying *in situ* arranged in a linear series, each unit closely following the other. The most distal of the isolated segments may continue into the narrow portion of Henle's loop (fig. 58).

In the dissection of these structures the greatest care was taken not to fracture a hypertrophic tubule and so produce artificial fragments. When isolated, each segment was carefully turned with the needles and examined at both ends for evidence of possible tearing or fracture. And to definitely establish that these objects are intact and independent structures, typical examples were removed from the suspending fluid, mounted on a slide beneath a cover slip and examined under high magnification. The pressure of the cover slip distorts their shape but has the advantage of flattening their contours. A photomicrograph shows that they are intact vesicles (fig. 63).

It is easy to see how such vesicles were unrecognized in histologic sections. Their contours are identical with those of the intact tubules among which they lie, and their epithelium, as can be seen by the thickness of their wall, is not atrophied, so that a section passing through them may differ in no essential way from one through a hyperplastic tubule that has maintained its normal continuity and that passes in and out of the section on its undulating course. We believe that even with serial sections one could decide only with considerable uncertainty whether or not a linear series of vesicles were connected.

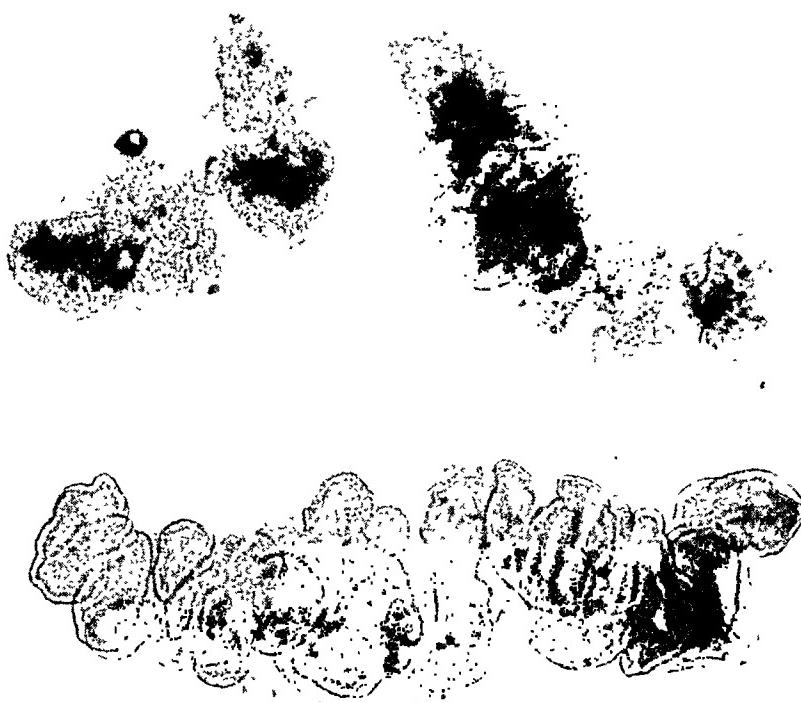


Fig. 63.—Three isolated vesicles similar to those shown in figure 61. The vesicles are shown by transmitted light at high magnification. Note that they are not artificial fragments but intact structures.

Since we have observed the effect of disruption at the junction of glomerulus and tubule and have seen the result of multiple interruption of the latter, it follows that the possibility remains of a single interruption occurring at some point in the tubule's course and this distal detached portion persisting more or less morphologically intact and unaffected. Such a structure would constitute an agglomerular tubule.

In the first study of this series a virtual agglomerular tubule was described and the significance of its occurrence noted. A fibrosed and therefore functionally absent glomerulus was attached to a hypertrophic tubule rather than to the collapsed and atrophied structure that has been presumed to follow glomerular obliteration. At the present time we

shall note only the occurrence of completely detached tubules; a detailed consideration of their structure with their relations to the detached glomerulus and blood vessels as illustrated by examples of actual isolated tubules and by reconstructions in models will comprise the third article of the present series. It is sufficient therefore to state that among the hypertrophied and hyperplastic terminal portions of the proximal convolution one frequently finds examples of these tubular segments completely cut off from their more proximal portion. The distal portions are morphologically well preserved in all detail and may continue uninterrupted, showing no unusual distortion or irregularity in their course (fig. 58).

#### COMMENT

If one considers the infinite modifications that are possible when pathologic processes including all the phenomena of degeneration, hyperplasia, hypertrophy and inflammation act over the course of years on a unit of such anatomic complexity as the nephron, it becomes evident that our description of the results of this first application of the method of microdissection to the problem of abnormal renal architecture makes no pretense at completeness. We believe that the more important of the typical alterations that occur in the nephron as a result of hemorrhagic Bright's disease have been described. Further study will supplement these observations, especially when the topographic relations of the abnormal nephrons are examined in the "contracted kidney."

Already certain new relations developing in the structure of the kidney as an organ have become apparent, for example, the mechanism of the shift in the circulation which long has been recognized, more, however, because its occurrence was demanded by logical considerations than because of exact objective demonstration. It is easily recognized by dissection that the isolated efferent vessels from the glomeruli are decreased in size, while Ludwig's vessels, normally so obscure that few observers have seen them, are found to be prominent structures as large as the afferent arteriole. The actual pathway of the blood stream as it is shunted past the obliterated capillary bed of the glomerulus can therefore readily be demonstrated. A further consideration of the details of these alterations in the blood flow will be given in the following article of this series in which the changes that occur in the tubule after glomerular obliteration will be described.

Another structural change in the abnormal organ is the occurrence of nephrons of a type not normally found in the human kidney. This change concerns the situation of the bend of Henle's loop, which always lies in the broad ascending segment in the short loops of the normal human kidney, though in the rabbit short loops may be found in which the bend is situated in the narrow segment. We have seen that when extreme hyperplasia occurs in the terminal portion of the proximal

convolution this structure not only kinks on itself as its cells increase in size and number, but also extends down deeper into the medulla. The thin segment of Henle's loop that formerly preceded it may be actually passed by the growing terminal segment of the proximal convolution and in fact be drawn after it as the hyperplastic tube advances. Consequently, the bend of the loop now lies in the narrow limb as in the nephron of the rabbit, and in some examples is situated at the immediate end of the proximal convolution.

Even more important general structural alterations result from tubular disruption, but a consideration of these will be reserved for the end of this section.

In the past there has been an inclination to think of the changes that occur in the nephron in Bright's disease as the result of a unit responding to general abnormal conditions. The tubule as a whole, or at least a complete division of it, is said to collapse when the glomerulus is destroyed, to hypertrophy when certain functional demands are thrown on it or to dilate when obstructed, or its epithelium is thought to undergo atrophy, degeneration, or death when adversely affected. Our examination of the entire nephron shows that this is not necessarily so. Fatty degeneration is frequently seen in the contiguous coils of a tubule cluster and these, when straightened out, show that only short segments, separated by normal-appearing epithelium, are involved. It is obviously a sharply localized factor that operated in such an instance, perhaps anoxemia due to the obliteration of a minute vessel, for no such effect is noted in the tubule as a whole. Similarly localized effects resulting from the pressure of connective tissue are seen alternating in the course of a single nephron. For example a large glomerulus is followed by an atrophic tubule which, as it passes to another region of the kidney, becomes hypertrophied and hyperplastic. Then extending to another area it returns to normal caliber and appearance, finally dilates and may repeat again almost any conceivable train of variation in its extended course through the diseased organ. In such cases one cannot speak of an "atrophic tubule" or of a "hypertrophied tubule." And since it is the nephron as a complete entity that is responsible for proper and complete function, the complications introduced in attempted correlation of structure and function by this variation within the unit are obvious and disconcerting. But a consideration of these difficulties can at least be postponed until the topographic problem of the abnormal kidney has been examined.

The importance of the proximal convolution is well established in the modern theory of renal activity. The observations in this study emphasize its paramount significance in the abnormal organ.

Only in the proximal convolution were progressive changes definitely observed. In this segment hypertrophy and hyperplasia were marked,

in fact, extreme, while all the other divisions of the tubule showed only changes of a passive or regressive nature, such as dilatation or fatty degeneration.

Especially prominent is the alteration seen in its terminal portion, the "spiral" of Schachowa and Schweigger-Seidel, an observation which correlates interestingly with the experimental deductions that from Ribbert and Susuki down to Richards in the present day suggest that various portions of the proximal convolution may have varying functions. So resistant, and, if one dare venture the word, so essential are these terminal portions of the proximal convolution that they persist as recognizable units even in the midst of the most disruptive of the disease processes. Vesicular structures and agglomerular tubules derived from them, still presenting evidences of vitality and progressive alteration, will be considered later.

The increase in length of the proximal convolution by the development in its terminal portion of hyperplastic kinkings and actual growth extension is a factor which has not previously been adequately recognized. In a previous study<sup>1</sup> the length of that portion of it which lies in the periglomerular cluster of loops was found by actual measurement of a reconstructed example to be 1.7 times the length of an average proximal convolution of the normal kidney. It was estimated therefore that, considering volumes, a hypertrophic unit of the terminal stage of the disease might replace, in physical size at least, six normal units. This estimate now needs correction, for the degree of the hyperplastic change in the terminal portion of the convolution was not recognized at that time. We now find that the hyperplastic terminal spiral may be equal in length to the periglomerular clustered portion, so that our former figure compared only one half of a possible hypertrophied proximal segment to the figure given by Peter for the entire normal convolution. The original length in our model of a complete hypertrophied convolution of 2.46 cm. may therefore be increased to 4.92 cm., and such an increase in length would double the volume. Our original statement should be amended to read that the hypertrophied unit may replace in physical size twelve normal structures. A considerable addition to the bulk of the persisting renal parenchyma is thus made by the hypertrophic and hyperplastic processes in the terminal portion of the proximal convolution.

The degree of hypertrophy and hyperplasia that have been noted in the proximal convolution and particularly in its terminal portion is a striking phenomenon, for nothing approaching the complex structural change that they produce has ever been noted in experimental studies of compensatory renal hypertrophy.<sup>10</sup> An explanation for this

10. Oliver, J.: Arch. Int. Med. 34:258, 1924. Peters, E.: Ztschr. f. Zellforsch. u. mikr. Anat. 8:63, 1928.

fact may possibly be found in the difference in the nature of the blood that supplies the tubules in a normal kidney undergoing compensatory hypertrophy after unilateral nephrectomy and that which circulates through the kidney in the terminal stage of hemorrhagic Bright's disease.

In the former case, the proximal convolutions of the tubule that are undergoing hypertrophy receive blood that has passed through the glomerulus and has therefore undergone whatever changes of elimination normally occur in this passage. Moreover, the glomeruli increase in size and presumably become more effective. The added burden that produces the hypertrophy of the tubule cells is therefore contained in blood that has been at least partially purified by the normal glomerular eliminatory procedure. In this instance tubular hypertrophy may be looked on as largely compensatory for tubular inadequacy.

In terminal Bright's disease, however, the greater part of the blood that reaches the proximal convolution has not undergone change by a passage through the glomerular mechanism, for it has been shunted directly by Ludwig's vessel to the tubule cells. Consequently they are burdened not only with products that have accumulated because of the destruction of other tubules, but by the added demand that results from glomerular destruction and insufficiency. In this instance tubular hypertrophy is compensatory for both tubular and glomerular inadequacy, and it might therefore be expected to assume unusual proportions. Further and even more direct evidence of the ability of the tubules to take over the function of the progressively decreasing glomeruli will appear in our next study.

Of equal importance with those active processes in the nephron that transfer waste substances from the blood into the lumen is the passive action of drainage by which the lumen is cleared of the products of elimination. The present study emphasizes the impedance to this drainage that develops in the abnormal kidney, for it illustrates the frequency and extent of the interference and shows the exact method and points in the nephron where obstruction is most apt to occur. It must be remembered that the only kidneys available for study were those that had failed completely, since they were selected from the terminal stage of the disease. The failing circulation and the consequent diminution in the flow of urine in the last few hours of life doubtless exaggerated some of the appearances noted, but other evidences are seen of obstruction of such long standing as to have caused permanent structural change in the tissues.

The simplest obstructing mechanism observed was the plugging of the lumen of the nephron with solid matter. Two distinct kinds of occluding material were observed, and these under conditions that indicate an entirely different significance. One substance is homogeneously

clear and translucent; the other is a heterogeneous mixture of granular detritus of varying density that is often mixed with droplets of fat. Associated with obstruction by either sort of material is found dilatation of the tubule, an occurrence which precludes the possibility that either might be a simple precipitation from albuminous urine by the action of the solution of formaldehyde or the acid. If in certain cases the dilatation is only moderate, it must be remembered that one of the well established functions of the tubule is the absorption of water, and this would be favored by the increase in pressure produced by the occlusion of the lumen.

The heterogeneous fatty granular material has all the appearance of débris that has been swept by the current of the urine from higher levels of the nephron to collect in certain traplike parts of the course of the tubule. It is found at tortuous or kinked points, particularly in the distal convolution and in the loops of the peripheral or in the abnormal deformities of the central collecting tubules. When found elsewhere it can be followed along the course of the tubule more or less continuously to one of these occluded points, an appearance which indicates that it has extended back by accumulation. This interpretation of an accumulation over a period of time is further supported by the stuffed appearance of the tubule with its irregularly bulging local dilatations and by the laminations and irregularities of density and of fatty admixture that build up the contents of the occluded tubule.

The clear translucent substance, on the other hand, is never limited to a definite restricted segment of the tubule but extends through long stretches, at times including an entire peripheral collecting tubule, the connecting piece, the distal convolution and the broad limb of Henle's loop. The entire length of the tubule is greatly and uniformly dilated, and its contours are rounded by an evenly distributed internal distention. The appearance is that of a marked dilatation with fluid that has collected behind an obstruction and then coagulated *en masse* at a given time. The actual obstruction may be the granular débris previously described. With such an interpretation the time of coagulation may have been either ante mortem or post mortem without affecting the significance of the result, since the obstruction is the essential matter.

None of the solid masses so far described comprise the casts that are found in the bladder urine. Obviously no mold of the lumen of the proximal convolution that approximates in diameter that of the typical urinary cast could pass through the narrow limb of Henle's loop, nor does it seem likely that one from the ascending limb could make its way through the complex and acutely bent kinks of even a normal distal convolution. This division in itself contains no sufficiently straight portions in which a urinary cast could be formed, and, as a matter of fact, no typically formed casts are ever seen in these upper reaches of

the tubule. It follows therefore that all those found in the bladder urine must be derived from the peripheral and central collecting tubules, though the material from which they are formed, whether débris, albuminous substances, desquamated epithelial cells, leukocytes or red blood cells, may be derived from any portion of the tubule, provided its lumen is not obliterated.

Typical straight well formed hyaline or granular casts with rounded ends were seen in all the collecting tubules. Since the course of these is direct and their lumens increase in size with their progression through a kidney the structure of which has not been greatly altered, their passage into the bladder urine is a simple matter. The only possible obstructions are the loops that occasionally occur in the course of the peripheral collecting tubules.

In the kidney whose structure has been altered, however, the fusiform segmented deformity so frequently observed in the central collecting tubules prevents the further passage of casts toward the pelvis of the kidney. As such deformities increase in number with the progress of the disease, and as heterogeneous débris clogs the irregularities of the larger ducts, the number of casts from the smaller peripheral collecting tubules must decrease. Only those formed in the terminal central ducts can enter the bladder urine. And in this fact a simple explanation is found for a clinical observation that has previously been obscure. In the last period of the terminal stage of the disease Addis has noted an increase in the size of the casts found in the urine.<sup>11</sup> Both the number of the larger casts and their average diameter increase progressively until their size approximates that of the ducts of Bellini. In the "cast count" of such a case the large renal failure casts rise to 100 per cent, while the percentage of hyaline and other casts of usual diameter falls to 0. It is evident that what occurs is a progressive clogging of ever larger collecting tubules, and that although this obstruction allows urine to filter through, it holds back the formed casts from the smaller tubes.

Obstruction affects in no small way the course of the structural changes in the terminal stage of the disease, and the final effects of many of these alterations are determined almost entirely by such "hydro-nephrotic" factors. And such effects are cumulative, for a vicious circle is soon established in which clogging and obstruction, as they produce stasis in the flow of urine, favor increased accumulation of débris. The permanency of such obstruction of the tubule by débris is hard to determine, but the inert substances as they are seen in the densely packed rigidly deformed distal convolutions and in the stuffed segments of the larger fusiform collecting tubules have an appearance of remarkable solidity and finality. Urine must be seeping past or through the occlud-

11. Addis, T.: J. A. M. A. 84:1013, 1925.

ing material, and it may be that in this way there results a certain disintegration of the distal end of the occluding mass, but a never ending source of supply for new accumulation is always present in the degenerating nephron above.

Interference with proper drainage is the obvious functional disturbance produced by the obstructive accumulations. That the more complex functional activities of the organ are affected in a deleterious manner seems equally certain, even if the exact nature of these disturbances is less clear. Back pressure would militate against excretion of both water and solids, whether they are eliminated by filtration or by secretion, while absorption of water or substances like urea might be exaggerated.

Obstruction to the flow through the nephron may also follow an actual interruption of the tubule by the pathologic reactions of the disease, and a result of these disruptions is the production of structures that have no analogies in the normal kidney. When the tubule is surrounded by inflammatory tissue it is compressed, and in the late stages when a dense collagenous scar is forming it may ultimately be pinched off completely. A nephron ending blindly is thus formed, which dilates with its accumulating excretion. Such an interruption may occur at any point of the nephron and is irreparable. Repeated interruptions of this kind also are found in the course of a single nephron. When the hypertrophic and hyperplastic terminal portion of the proximal convolution is involved a series of completely closed vesicles is formed the irregular contours of which retain the characteristic appearance of the original hyperplastic tubule.

Even more frequent is the multiple interruption of tubules of normal caliber. Since an area of scar tissue may be extensive, a great number of tubules may be thus included throughout a considerable stretch of their course, and the final effect is a mass of clustered small round cysts the diameters of which are equal to that of the original tubules.

Such vesicles and cysts are, of course, functionless and in certain cases may comprise a considerable part of the epithelial-tubular element of the diseased organ. Their recognition is therefore important when any attempt is made at correlation of structural change and functional ability. In the ordinary histologic preparation, however, both vesicles and cysts have in cross-section an appearance almost identical with that of the tubule from which they are derived. In looking over our earlier histologic descriptions of these same kidneys we have found these worthless cysts described as "tubules." Such an interpretation, whatever functional activity might have been assigned to the tubules, considers them at least capable of transporting urine. In the light of this dis-

crepancy, it is not surprising that correlation of structure and function has been inadequate when, as in the past, it was made solely by the study of sections.

Another result of the disruption of the nephron is concerned with the persistence of portions of the proximal convolution as independent structures which do not of necessity atrophy or collapse. They have been found not only to persist but to show the active progressive changes of hypertrophy and hyperplasia. They constitute in a very strict sense agglomerular tubules and will form the subject of the next study of this series.

LIGATION OF THE COMMON BILE DUCT IN THE RAT  
ANATOMIC AND BEHAVIORISTIC EFFECTS

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Previous papers from this laboratory have reported experiments in which an attempt was made to study the rôle played by various organs of the body in the production and regulation of spontaneous activity. In brief, the object of these experiments was to determine what makes an animal active, how it can be made chronically overactive (experimental mania), how it can be made chronically inactive (experimental depression) and finally how it can be brought back to normal again, once these abnormal states of activity have been produced. It is hoped that with knowledge thus obtained of the principles underlying activity in animals, some light may be thrown on the origin and control of abnormal states of activity in man.

One approach to this problem has consisted of an attempt to determine the effect produced on activity by removal of various organs of the body. In this way it has been found that the endocrine glands play a particularly important rôle, since removal of the gonads, hypophysis, suprarenals or thyroid results in a great permanent drop in activity. In marked contrast, removal of the pineal gland, salivary glands, thymus, spleen, uterus, seminal vesicles or prostate produces no change in activity (Richter<sup>1</sup>).

As a part of this survey we have investigated the liver, because of its recognized function of regulating metabolism and the general energies of the body. Owing to technical and vital physiologic difficulties it was thought best not to extirpate the liver but rather to be content at the outset with a study of the effects produced on activity by a marked alteration in its function, leaving it *in situ*. The most obvious procedure was to study first of all the effect produced by ligation or actual transection of the common bile duct. By this operation the channel through which the secretions from the liver pour into the intestinal tract is closed, while the connection between the liver and the blood stream is still intact.

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1. (a) Richter, C. P.: Quart. Rev. Biol. **2**:307, 1927; (b) Am. J. Orthopsychiat. **2**:345, 1932.

These experiments have brought out certain relations between the liver and spontaneous activity, but of even greater interest are the amazing anatomic changes produced by ligation of the bile duct, not only in the liver but also in the bile duct itself.

#### METHODS

The technic followed in measuring the activity of the rats has been described in detail in a previous paper (Richter<sup>1a</sup>). Each animal was kept in a separate cage containing a revolving drum and a living compartment, with a food cup and water bottle. The animals had free access to the revolving drum, the revolutions of which were recorded on a cyclometer. Activity and the intake of food and water were recorded daily and the animals were weighed once each week.

Twenty-four animals were used in these experiments. They were placed in the cages at the usual age, between 35 and 45 days, but the bile ducts were not ligated until several weeks later, when a fairly high running level, approximately 6,000 to 8,000 revolutions per day, was attained. The age at the time of the operation varied from 41 to 164 days.

The operative technic was simple. Under ether anesthesia a 2 inch (5 cm.) midline incision was made in the abdominal wall beginning just below the sternum and exposing the liver and stomach. It was found that the exposure could be greatly improved by placing silver retractors so that they pulled the walls slightly upward as well as outward. A small silver retractor covered with cotton was inserted into the incision from the anterior end and hooked under the liver, so as to make an opening between the liver and the stomach and thus expose the common bile duct clearly to view.

In some of the animals the duct was tied with a single silk ligature and left unsevered; in others it was tied off with a double ligature and transected between.

At autopsy the abdominal organs were subjected to careful gross examination, and the glands of internal secretion and the brains were removed for histologic study. The liver was fixed in a diluted solution of formaldehyde, U. S. P. (1:10) embedded in paraffin, sectioned at 10 microns and stained with hematoxylin and eosin.

#### RESULTS

*Effect on Activity.*—Ten of the twenty-four animals became markedly inactive after the ligation of the common bile duct (group I). The remaining fourteen either showed no effect, or, after a short period of inactivity, became as active as normal animals and, in a few instances, even more active (group II).

Two typical records from animals of the inactive group are presented in figure 1, in which the running activity, expressed in the number of revolutions of the drum, is indicated on the ordinates and the age of the animal in days on the abscissas. The animal represented at the left became totally inactive after the bile duct was ligated, its activity dropping from a level of over 8,000 revolutions per day to less than 100. Death occurred fifty days later. The animal represented at the right also was totally inactive at the time it died, fifty-four days after the ligation of the duct. In contrast to the first animal, however, it remained fairly active for fifteen days after the ligature was applied.

### Daily Running Activity

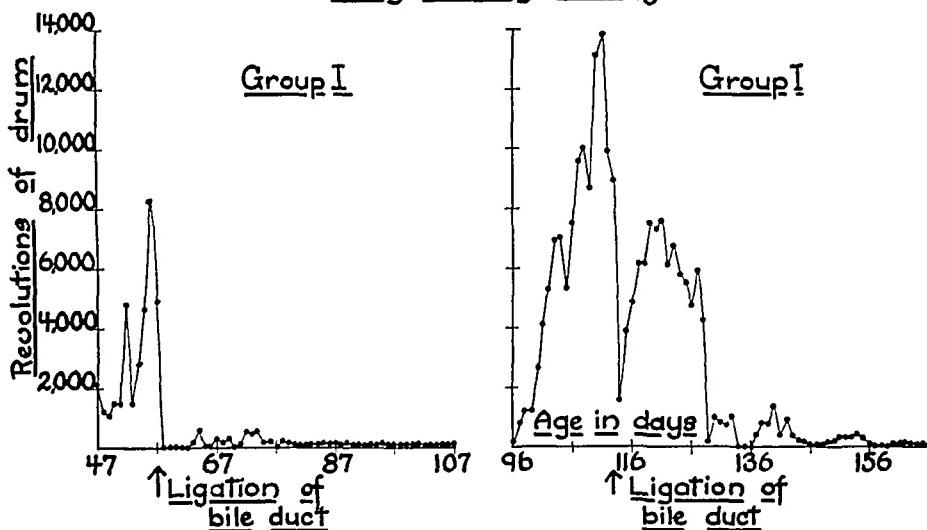


Fig. 1.—Effect produced by ligation and cutting of the bile duct on the running activity of animals in group I.

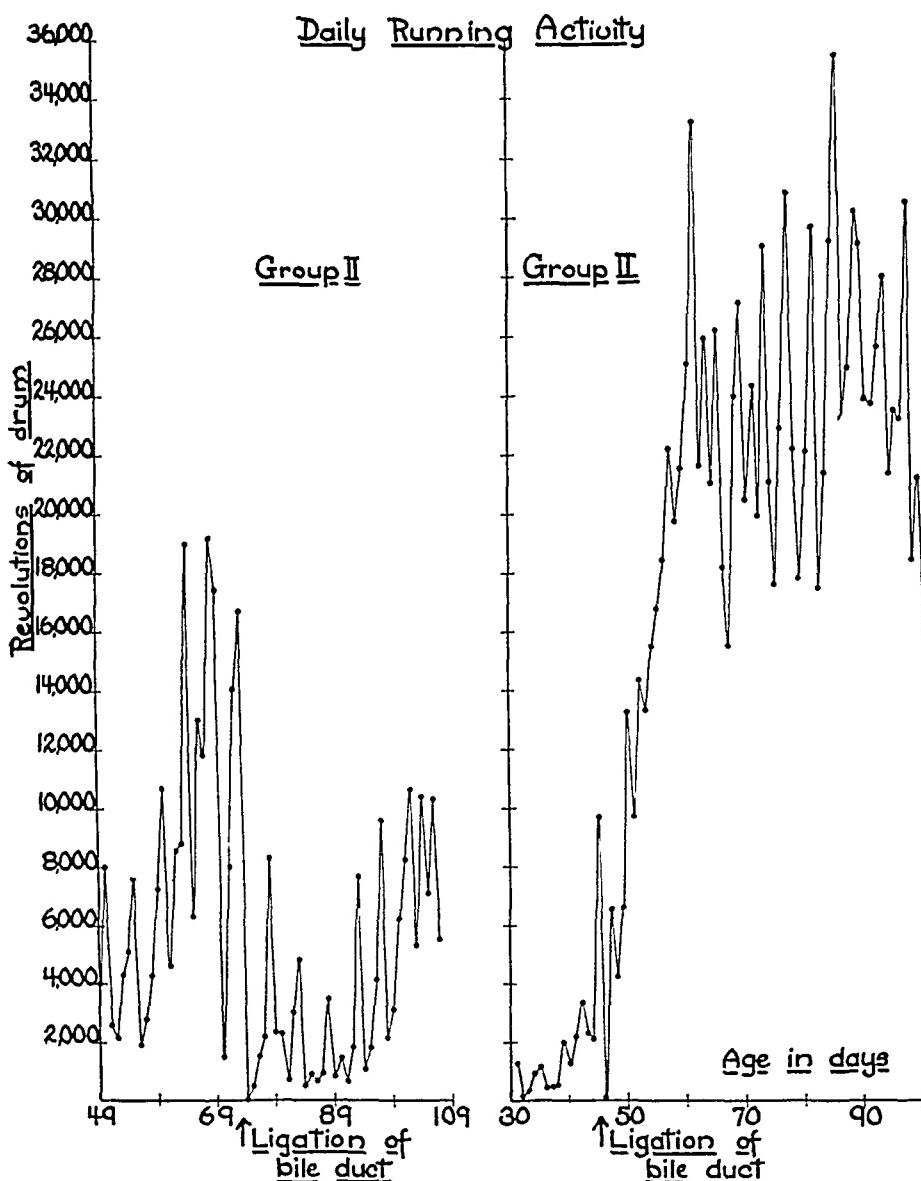


Fig. 2.—Effect produced by simple ligation of the bile duct on the running activity of animals in group II.

Experimental Records of Animals in Groups I and II

Rat Group I	Sex	Age at Operation, Days	Length of Life after Operation, Days	Position of Ligature	Average Activity* Ten Days Last Before Five Days Operation of Life	Dilatation of Common Bile Duct	Degree of Biliary Chrosis		Proliferation of Bile Canaluli		Degree of Fibrosis	Bile Duct	Status of Pancreas on Gross Examina- tion
							8	7	9	10	11	12	13
118C	F	37	55 (died)	Double ligature and cut	2,971	13	Entirely gone	Very marked	Very slight	Very marked	Slight	Not patent	Atrophied
119C	M	138	16 (died)	Double ligature and cut	5,037	493	Entirely gone	Very marked	Very marked	Very marked	Slight	Not patent	Slightly atrophied
20C	F	86	20 (killed)	Double ligature and cut	11,467	635	Entirely gone	Very marked	Very slight	Very marked	Very slight	Not patent	Markedly atrophied
329B	F	114	39 (killed)	Single ligature near liver	4,895	4	Entirely gone	Very marked	Very slight	Very marked	Slight	Not patent	Atrophied
315B	F	114	55 (killed)	Single ligature near liver	9,217	24	Entirely gone	Very marked	Very marked	Very marked	Very slight	Not patent	Markedly atrophied
282B	F	164	44 (died)	Single ligature near liver	14,100	197	Entirely gone	Very marked	Very marked	Very marked	Very slight	Not patent	Markedly atrophied
127C	F	11	40 (died)	Double ligature and cut	8,346	569	Markedly decreased	Slight	Marked	Very slight	Very slight	Not patent	Normal
374B	F	51	31 (killed)	Single ligature near duodenum	7,771	1,964	Entirely gone	Very marked	Very slight	Very marked	Slight	Not patent	Markedly atrophied
123B	F	14	29 (died)	Double ligature and cut	1,915	431	Entirely gone	Very marked	Very marked	Very marked	Moder- ate	Not patent	Markedly atrophied
144B	F	47	35 (died)	Double ligature and cut	6,214	1,181	Entirely gone	Very marked	Marked	Marked	Slight	Not patent	Atrophied
Average (for 10 rats)		37			7,193	551							

Experimental Records of Animals in Groups I and II—Continued

\* Expressed in number of revolutions on the drum.

The curves in figure 2 were obtained from the active group. The animal whose record is shown at the left was inactive for about ten days after the operation but was rapidly approaching the original high level of activity when it was killed thirty-four days afterward. The animal whose record is presented at the right showed no initial depression after the ligature of the bile duct, but became more active at once, finally reaching a level of about 28,000 revolutions per day, which is considerably above the normal average. It was killed fifty-five days after the operation.

The results of these experiments are summarized in the table, the scheme of which is self-evident. The records for the ten animals of group I are given at the top of the table, those for the fourteen animals of group II below.

In the sixth and seventh columns it will be seen that the average activity of group I dropped from 7,193 revolutions per day to 551, while that of group II increased from 8,069 revolutions per day to 8,874. An inspection of the individual records of the animals of group I shows, moreover, that every animal became much less active after the ligature of the bile duct. One animal averaged only 4 revolutions per day, whereas some of the animals of group II averaged as high as from 16,000 to 18,000 revolutions.

It will be noted in the fourth column that the six animals of group I that were not killed died from sixteen to fifty-five days after the ligature was made, or after an average of thirty-six days, whereas none of the animals of group II died. There is little reason to doubt that the other four animals in group I would have died at about that time had they not been killed.

The fifth column discloses the noteworthy fact that all of the six animals in which the duct was cut as well as ligated fall into group I. Otherwise, the type and position of the ligature, whether single or double and whether nearer the liver or nearer the duodenum, seem of no importance in separating the groups.

*Effect on the Liver.*—At autopsy marked organic differences, both macroscopic and microscopic, were observed in the animals of the two groups. These data are summarized in columns 8 to 15 inclusive of the table.

In all of the animals of group I a marked dilatation of the common bile duct was noted. This duct, which in the normal animal is scarcely larger than a small thread, had dilated so much that in some cases it almost filled the entire abdominal cavity (fig. 3). Three of the animals of group II showed a very slight dilatation of the duct, the others none.

The patency of the bile duct was tested by means of injections of india ink made at the point at which the bile duct enters the duodenum and at which it leaves the liver as the common duct. It was noted that

in nine of the animals of group II the ink went through the duct, whereas in all of the animals with the marked dilatation (group I) it did not.

The retroperitoneal fat had entirely disappeared in all but one animal of group I and in this it was markedly diminished. In group II, on the other hand, the deposit of fat was essentially normal. Histologic study of sections of the liver revealed a striking biliary cirrhosis with some necrosis, marked proliferation of the bile ducts and general diffuse

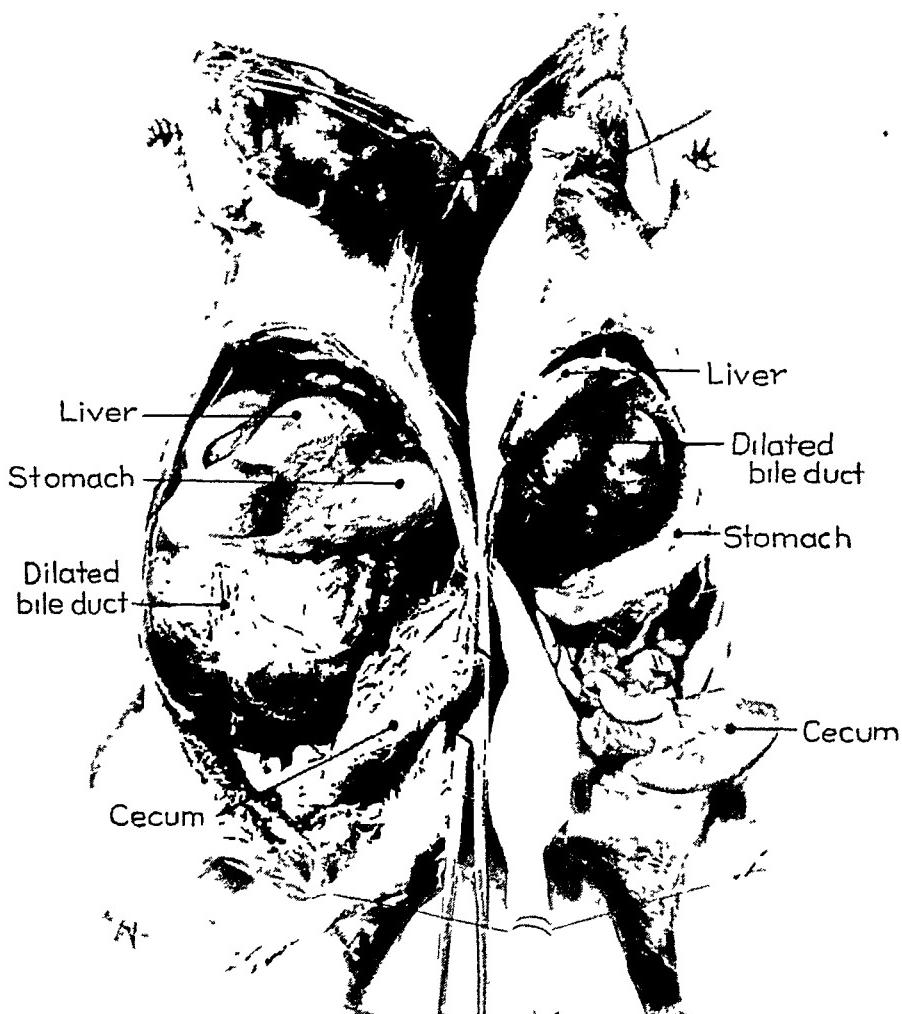


Fig. 3.—Two rats with markedly distended bile ducts.

periductal fibrosis (columns 10 to 14 in the table). In the animals of group II there were only very slight cirrhosis and necrosis and slight proliferation of the bile duct and fibrosis.

In the fifteenth column of the table it will be seen that the pancreas was atrophied in all but one of the ten animals in group I, and in only one of the animals of group II. In several cases the atrophy was striking.

In summary it may be said that in the animals of group I made inactive by successful ligation of the bile duct, a damming up of bile caused a marked dilatation of the common duct, with widespread destruction of liver cells, marked cirrhosis and proliferation of the small bile canaliculi or ducts together with a complete disappearance of the retro-peritoneal fat and an atrophy of the pancreas. In the animals of group II, with an unchanged or heightened curve of activity, the secretion of bile escaped through the normal channels provided by the regenerated bile duct. The slight alteration in the liver noted in these animals must have been produced during the short interval after the ligation and before the new channel had been formed.

These results confirm the observations of Cameron and Oakley,<sup>2</sup> in 1932, to which our attention has recently been drawn. The changes described in the present series are somewhat more pronounced, however, since the dilatation of the bile duct was greater and the alteration in the liver more marked.

#### COMMENT

These experiments were undertaken to determine whether changes in the liver resulting from ligation of the bile duct have any effect on spontaneous activity. The purposes of the experiment were defeated in over half of the animals by the unexpected regeneration of the bile duct, and in the others the conditions were much complicated by the appearance of the large dilatation of the bile duct, which in itself might have a profound effect on activity. Thus it became difficult to determine to what extent the marked inactivity of the animals in the latter group could be ascribed to the changes in the liver following the blocking off of the normal channels of escape of the bile, or to the mechanical pressure exerted by this large sac on the various internal organs as well as on the general circulatory relationships.

The bulk of the evidence favors the view that the changes in the liver with the probable presence of bile salts and acids in the circulating blood were the primary factors, since in almost every instance the animal became inactive immediately or within a few days after the ligation, long before the bile duct had reached an appreciable size.

An attempt was made in four animals to settle this question by means of a fistula of the bile duct. The idea was to determine whether animals made inactive by ligation of the bile duct become normally active again by permitting the bile in the sac to drain off freely to the outside through a fistula. The operative difficulties did not make it possible to settle this question. All of the animals died of abdominal hemorrhages resulting from the release of pressure after the saccular bile duct was opened to produce the fistula. It may be mentioned that particular care was used to make

2. Cameron, G. R., and Oakley, C. L.: J. Path. & Bact. 35:769, 1932.

the escape of fluid very gradual in order to avoid a sudden drop in abdominal pressure. The slight release in pressure from a small incision in the abdominal wall apparently is sufficient to produce fatal internal hemorrhages. However, as before stated, the rest of the evidence seems to indicate that the inactivity is not produced by the dilatation but by the changes in the liver.

The nature of the mechanism through which this inactivity is produced is not clear. It seems likely, however, that the damming up of the bile and its consequent forced injection into the blood stream in abnormally large quantities may be an important factor. This is suggested particularly because of the work of Crandall and Weil<sup>3</sup> in which it was shown that the bile acids had a destructive action on the nerve tissue in rats, and further that ligation of the common bile duct in dogs produced widespread atrophy of the tissue of the brain, particularly of the lenticular nucleus. Such great destructive changes in the brain tissue might easily account for the marked inactivity. However, against this possibility several facts can be offered: First, the rats do not show definite jaundice, except when the bile duct ruptures, and second, the animals become inactive within the first few days after the ligation, whereas, according to Crandall and Weil, no histologic changes appear in the brain of the rat until from four to eight weeks later. In agreement we noted no detectable histologic changes in the brains of six of our animals killed three weeks after the ligation of the bile duct.

We hope to be able to throw light on this problem by further experimentation on the effect produced on activity by injection or feeding of bile acids and salts.

An attempt also was made to find some explanation for the increased activity of a few of the animals of group II in which the bile duct had regenerated after the suture. It occurred to us that in these animals the ligation might in some way have made it possible for the liver to retain or store more glycogen and consequently have more energy available for running activity. However, stains for glycogen on sections from the livers of these and normal animals showed no appreciable differences.

#### SUMMARY

In twenty-four rats the common bile duct was ligated singly or doubly and then cut, and a study made of the effect produced on spontaneous running activity. An attempt was made to correlate these effects with histologic changes in the liver.

Ten of the animals became very inactive; fourteen either remained normally active or possibly became even hyperactive.

3. Crandall, L. A., and Weil, A.: Arch. Neurol. & Psychiat. **29**:1066, 1933.

All of the inactive animals showed striking changes at autopsy: The common bile ducts were markedly dilated, in some instances practically filling the entire abdominal cavity; the retroperitoneal fat was absent, and the pancreas atrophied. Histologically the liver showed a typical biliary cirrhosis with extensive sprouting of small bile ducts and necrosis.

The active animals showed a patent duct with slight or no dilatation, normal retroperitoneal fat and a normal pancreas. Histologically the liver showed slight biliary cirrhosis and proliferation of the bile canaliculi.

It was pointed out that several factors must be taken into consideration in explaining the inactivity produced by ligation of the bile duct—the mechanical effects of the presence of the large sac in the abdominal cavity, its pressure on the other organs and the chemical effects of the bile injected into the blood stream and acting particularly on the brain.

# STUDIES IN ATHEROSCLEROSIS: CHEMICAL, EXPERIMENTAL AND MORPHOLOGIC

## V. POSSIBLE DANGERS OF IODINE THERAPY IN ATHEROSCLEROSIS OF AORTA SEEN FROM AN EXPERIMENTAL STANDPOINT

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CHICAGO

The supposed action of iodine in atherosclerosis of the aorta is that of (1) healing by direct influence on the atheroma or by indirect influence on the intermediary cholesterol metabolism (Liebig, Hildebrandt, Abelin, Friedland), (2) increasing the functional discharge of blood from vessels and thus maintaining a better supply of blood to the organ (Guggenheimer and Fisher), (3) lowering the blood pressure (Potain and Thoussig) and (4) reducing the viscosity of the blood (Müller and Inada, Romberg).

All of these activities have been open to question experimentally. Thus Mancke, also Freund and König could not verify the work of Guggenheimer and Fisher, who described a widening of the coronary arteries after transfusing hearts with diluted solutions of iodine; Gumprecht, Stockman and Charteris, and Janeway found no decrease in blood pressure, while Blum (cited by Liebig) found that the blood pressure might be lowered or elevated; Brokling and von Borutta (cited by Liebig) found no changes in the viscosity of the blood after the administration of iodine.

In support of the first hypothesis, Liebig was first to show that iodine in massive doses prevented atheromatous deposits in cholesterol-fed rabbits (later Turner), while Seel and Creuzberg reported that by using small doses of iodine not only could the deposit of lipoid in the aorta of the cholesterol-fed rabbit or guinea-pig be prevented, but after a steatosis had developed, it could be reversed by the administration of iodine.

The recent studies of Turner and Khayat show that iodine was not effective unless the thyroid gland was present. These authors allowed one to infer that the action of the iodine was an indirect one working through the thyroid gland.

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From the Pathologic Institute, Freiburg, Germany, Dr. L. Aschoff, director; the chemical division of the Pathologic Institute, Freiburg, Germany, Dr. Schönheimer, formerly director, and the Pathologic Institute of the Cook County Hospital Chicago, Dr. R. H. Jaffé, director.

That the thyroid gland played some rôle in the prevention of experimental atherosclerosis of the aorta was shown by Murata and Kataoka, who prevented this state by the administration of thyroxine, and verified by Friedland and more recently by Turner. On the contrary, the removal of the thyroid gland hastened the development of atheromatous deposits in the aorta (Shapiro, Friedland). Whether the influence of the thyroid gland prevents absorption of cholesterol or increases excretion of cholesterol has not been demonstrated, but it is definitely known that with large doses of iodine, thyroxine or whole thyroid, the blood cholesterol in the experimental animal is diminished (Liebig, Turner, Turner and Khayat). The degree of atherosclerosis of the aorta in experimental animals is directly proportional to the level of the blood cholesterol (Anitschkow).

Unfortunately, in the studies of the action of iodine reported, the thyroid gland was not examined nor was the basal metabolic rate, so that the degree of activity of the thyroid gland cannot be stated. In the studies of the action of thyroxine Friedland showed that a definite increase of the basal metabolic rate as well as a loss of weight occurs. Abelin found as much as a 100 per cent increase of the oxygen requirement in mice fed thyroxine. Microscopic examination of the aortas of the animals fed large doses of thyroxine (Friedland) or iodine (Hedinger and Lach, cited by Liebig) showed severe medial calcification similar to the epinephrine type of sclerosis.

From an experimental standpoint it is suggestive that the action of iodine is determined by the activity of the thyroid gland and that although the deposits of lipoid are hindered when massive doses of iodine or thyroxine are administered, a severe medial calcification follows.

The question arises: What will large physiologic doses of iodine do toward the prevention of deposits of lipoid in the aorta of the cholesterol-fed rabbit? This problem has been elaborated on and will be discussed in later paragraphs.

#### METHOD

Rabbits of about the same age (1 year) were used. Except for two of the controls, all were fed, on the average, 0.5 Gm. of cholesterol dissolved in 10 cc. of linseed oil daily for about one hundred and twenty days. This substance was administered by the aid of a stomach tube. The regular diet of hay, oats and later turnips and grass was given in addition to the cholesterol.

Studies were made on the following groups: Group 1 (controls—subgroups *a* and *b*, 5 animals each) fed (*a*) cholesterol in oil plus the regular diet; (*b*) regular diet only. Group 2 (subgroups *a* and *b*, 5 animals each) fed (*a*) cholesterol in oil plus potassium iodide (at first 3 mg. of potassium iodide, equal to 2.15 mg. of iodine per day,

which was increased to 4.5 mg. of potassium iodide, equal to 3.37 mg. of iodine per day); (b) cholesterol in oil to which potassium iodide was added six weeks later (doses of potassium iodide same as in a). Group 3 (subgroups *a* and *b*, 3 animals each) fed (*a*) cholesterol in oil plus Jodtropon<sup>1</sup> (40 mg. of Jodtropon, equal to 2 mg. of iodine per day, which was increased to 66.5 mg., equal to 3.37 mg. of iodine per day); (b) cholesterol in oil to which Jodtropon was added six weeks later (doses of Jodtropon the same as in *a*).

Determinations of the blood cholesterol, free and combined, were made weekly, employing a newly devised method of Schönheimer and Sperry (not yet published) in which very small quantities of serum were needed (0.2 cc.). The principle involved in this method is similar to that in Windaus' use of digitonin for the precipitation of the cholesterol, but the determination quantitatively of the digitonin cholesterol precipitate is made photometrically using anhydrous acetic acid and concentrated sulphuric acid for the color formation. As no other reports on iodine gave the relations of the free cholesterol to the ester in the blood and because only one other report on cholesterol feedings gave these relations and that one to a limited degree (Wacker and Hueck), a more detailed description of these will be given. The blood cholesterol curves of only one animal of each group are given. Although there were variations in each group, the curves given show the general trend of each division.

#### CHOLESTEROL CHANGES IN THE BLOOD SERUM

*Group 1 a* (regular diet plus cholesterol in oil).—The total serum cholesterol increased slowly from a normal of 55 to 65 mg. per hundred cubic centimeters to double that much in the third week, then it sank to a little above normal and remained constant until the fifth week, when a sharp rise occurred extending over a period of from three to four weeks with the values soaring to an average of 900 mg. per hundred cubic centimeters. From then on a sharp fall for about three weeks was noted, then again a rise, then a fall, and finally a gradual rise by a stepladder type of ascent to 1,400 mg. per hundred cubic centimeters.

The relationship of free cholesterol to cholesterol esters, which normally varies from 35:65 per cent to 25:75 per cent, attained a high normal value averaging 26:74 per cent. For short periods (one week) an increase of the cholesterol ester to 80 per cent or slightly over occurred (fig. 1).

*Group 2 a* (regular diet plus cholesterol plus potassium iodide).—The total serum cholesterol increased very rapidly and at the end of the third week reached on the average 500 mg. per hundred cubic centi-

1. Jodtropon is an organic iodine bound intramolecularly with protein. It is manufactured by the Troponworks Dinklage & Co., Cologne-Mühlheim, Germany.

meters. In the following week it fell to about twice the normal value, while in the next five weeks a very acute rise took place, reaching 2,500 mg. per hundred cubic centimeters (the low value was 1,600 mg.). A slight drop followed for a week or two and then again a high level was attained.

The percentage of cholesterol ester was somewhat above the high normal with an average of 78 per cent (fig. 2).

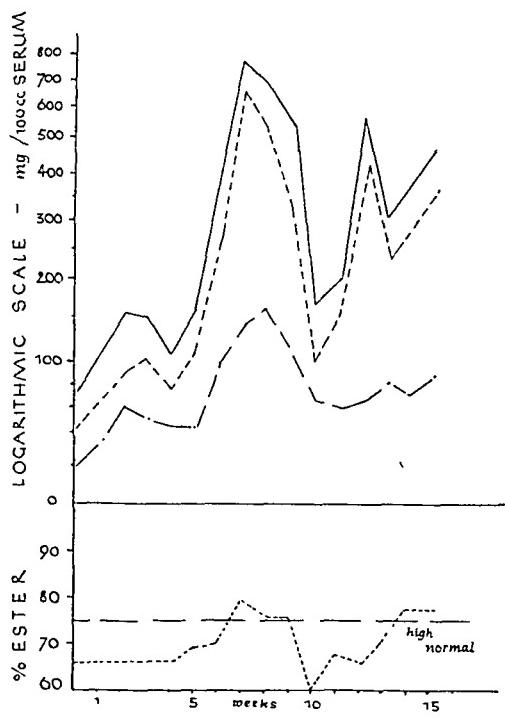


Figure 1

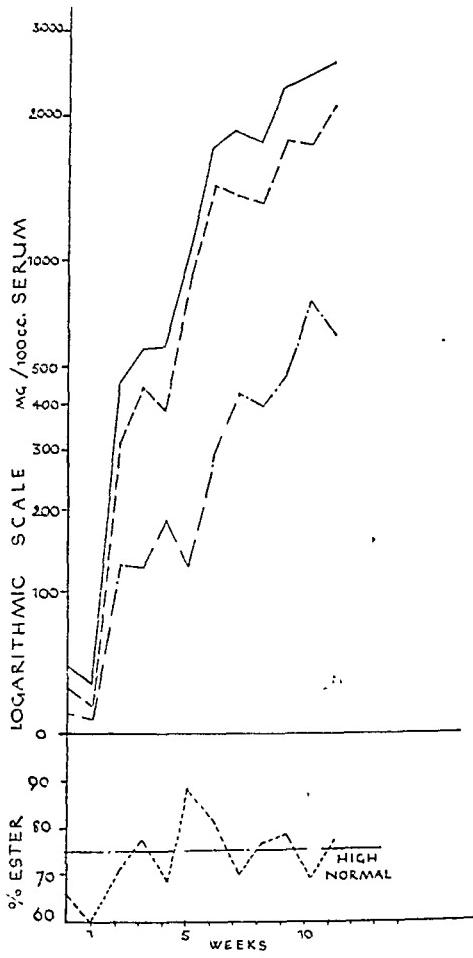


Figure 2

Fig. 1.—Cholesterol changes in the blood serum of a representative rabbit (83) of the control group 1a which had cholesterol in oil added to the regular diet. In all the charts the total cholesterol is represented by an unbroken line; cholesterol ester, by a line of dashes, and free cholesterol, by a line of dots and dashes. At the foot of all the charts, the changes in the percentage of the serum cholesterol ester are shown. The high normal level is indicated by a line of dots and dashes.

Fig. 2.—Cholesterol changes in the serum of a representative rabbit (87) of the group 2a given, in addition to the regular diet, cholesterol in oil and potassium iodide.

*Group 2 b* (regular diet plus cholesterol, with potassium iodide added after six weeks).—The total serum cholesterol curve was similar to that of group 1 *a* for the first six weeks. At this time, when usually a drop set in, potassium iodide was added, and the cholesterol value remained stationary for that week instead of falling. After that the rise was very abrupt, going steadily upward, in some cases reaching a high level of 2,700 mg. per hundred cubic centimeters (low value, 1,422 mg.).

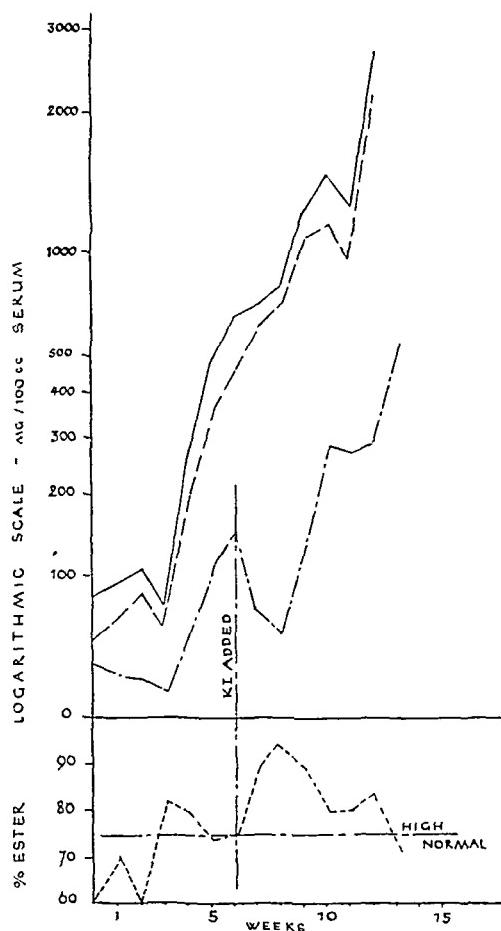


Fig. 3.—Cholesterol changes in the blood serum of a representative rabbit (26) in the group 3*b* given, in addition to the regular diet, cholesterol in oil to which six weeks later potassium iodide was added.

The relationship of cholesterol esters was definitely altered. Whereas for the period prior to the addition of potassium iodide cholesterol ester was 74 per cent, as found for all the rabbits to the diet of which cholesterol alone was added, after the administration of potassium iodide a sudden increase of the percentage occurred, reaching 90 and later 94. After six weeks, a gradual return to the high normal occurred. The average percentage of cholesterol ester after potassium iodide was added was 83 (fig. 3).

*Group 3a* (regular diet plus cholesterol plus Jodtropon).—The findings for Jodtropon were similar to those for potassium iodide (group 2a), except that the rise of the total serum cholesterol was more abrupt with a steady incline and only an occasional divergence. The highest level reached was 2,662 mg. per hundred cubic centimeters.

The cholesterol ester relationship was similar to that when potassium iodide was administered, viz., 78 per cent (fig. 4).



Figure 4

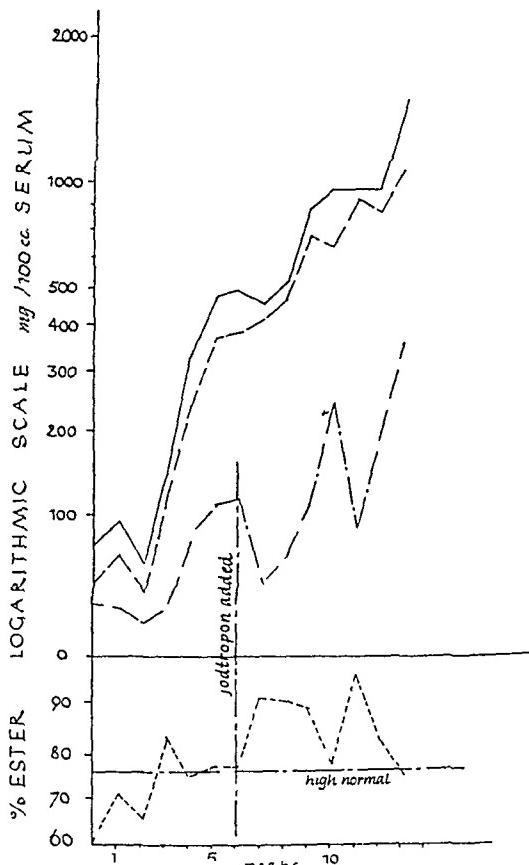


Figure 5

Fig. 4.—Cholesterol changes in the blood serum of a representative rabbit (29) in the group 3a given, in addition to the regular diet, cholesterol in oil and Jodtropon.

Fig. 5.—Cholesterol changes in the blood serum of a representative rabbit (27) in the group 3b given, in addition to the regular diet, cholesterol in oil to which Jodtropon was added six weeks later.

*Group 3b* (regular diet plus cholesterol, with Jodtropon added after six weeks).—The blood cholesterol findings were similar to those for group 2b, except for a slight drop of the total cholesterol in the six weeks.

The cholesterol ester relationship was 74 per cent before and 83 per cent after the administration of Jodtropon (fig. 5).<sup>1</sup>

*Summary.*—The serum cholesterol increased in all cases in which cholesterol in oil was fed. On the average, when cholesterol alone was added to the diet, the rise was slow in the first three weeks and then abrupt, reaching the high level of about 900 mg. per hundred cubic centimeters. When iodine in the form of potassium iodide or Jodtropon was fed along with cholesterol in oil, the rise in blood cholesterol was more acute and the levels reached were as high as 2,500 mg. per hundred cubic centimeters. If cholesterol in oil was first fed and then iodine added to the feeding, the course followed simulated the one taken when iodine was fed from the start, with similar high values. The sex of the animals made no appreciable difference to this relationship.

The cholesterol ester relationship reached high normal values (74 per cent) when cholesterol alone was added to the diet, and somewhat higher values when iodine was given simultaneously (78 per cent), but when cholesterol was fed and iodine added later, this relationship reached the high value of 94 per cent, with the average being 83 per cent. The cholesterol ester content of the blood remained high for six weeks, at which time it returned to normal.

In view of the possibility of an error of 5 per cent in the chemical technic employed, it may be conceded that the difference in percentage of cholesterol ester between the first two groups might become insignificant, but in the last group the difference is beyond the possibility of error.

#### AORTIC CHANGES

*Group 1 a* (cholesterol in oil added to diet).—There was a slight to moderate development of yellowish-gray, elevated plaques situated mostly about the mouths of the innominate, left subclavian, carotid and intercostal arteries.

The microscopic changes consisted of a subendothelial deposit of cholesterol ester, free and in histiocytes. There was a moderate amount of fat between the elastic fibers of the inner third of the media. This deposit was found as a rule along the distal aspects of the fibers. There was no elastic tissue proliferation but rather a spreading apart of the fibers.

*Groups 2 a and 3 a* (cholesterol and iodine added to diet).—The degree of lipoid deposit was more marked than in group 1. In addition to the deposit about the mouths of the vessels, there were plaques and long linear deposits along the posterior wall, not related to the intercostal arteries. To a lesser extent small plaques were found in the abdominal portion.

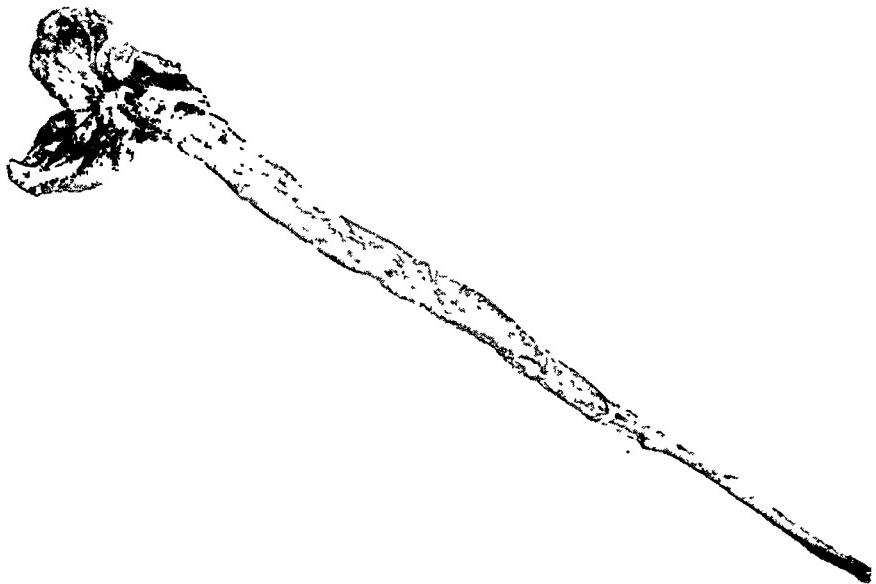


Fig. 6.—The aorta and heart of a rabbit fed, in addition to the regular diet, cholesterol in oil to which potassium iodide was added six weeks later (rabbit 26). The entire specimen was stained with sudan III. Note the marked lipoid deposit, especially in the thoracic portion (one half normal size).



Fig. 7.—Several thicknesses of the aorta stained with sudan III. This rabbit was fed, in addition to the regular diet, cholesterol in oil to which after six weeks Jodtropon was added (rabbit 27). Note the marked lipoid deposit extending into the media (dark-staining substance);  $\times 50$ .

The microscopic observations were similar to those in group 1 *a*, except that they were more pronounced. The intima became thickened by the lipoid deposit; there was a marked proliferation of histiocytes, and the elastic fibers of the inner media were spread far apart by the infiltrating lipoid. No elastic tissue proliferation was found.

*Groups 2 b and 3 b* (cholesterol added to the diet, with iodine preparations added after six weeks).—The aortas of this group showed the most marked changes. Figure 6 shows an aorta stained with sudan III in which practically the entire thoracic portion was infiltrated with fat, and to a lesser extent the abdominal portion.

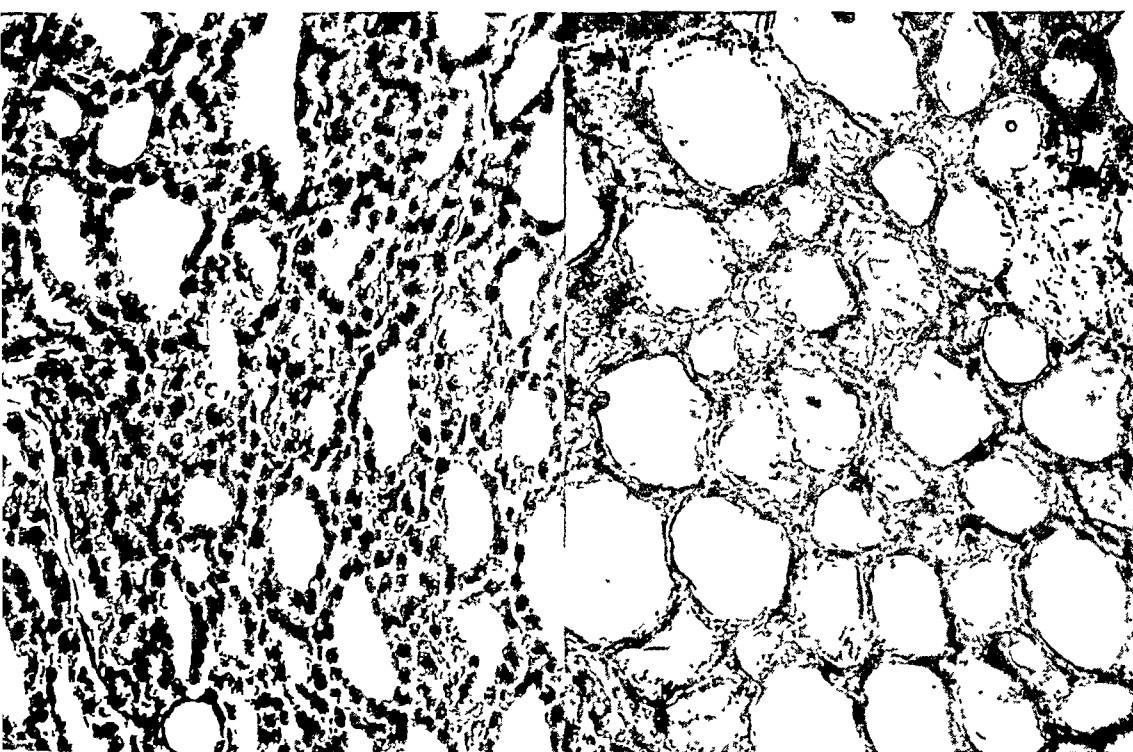


Fig. 8.—Left, thyroid gland of a rabbit which had cholesterol only added to its diet. Note the cuboidal epithelium and the small follicles. Right, thyroid gland of a rabbit which had both cholesterol and iodine added to its diet. Note the pavement epithelium and the large follicles (hematoxylin and eosin;  $\times 500$ ).

The microscopic changes are best seen in figure 7, which shows that the inner half of the aortic wall was infiltrated with fat. The intima was thickened either diffusely or in plaques by the infiltrating fat. No elastic tissue proliferation was found.

*Summary.*—In attempting to correlate the anatomic changes of the aorta with the blood findings it appeared that the amount of fat in the aorta was roughly proportional to the degree of hypercholesterolemia, and that of the latter the ester was the important component (Aschoff, Anitschkow, Schönheimer).

## CHANGES IN THE THYROID GLAND

In seeking to explain the action of the iodine, the thyroid glands were studied microscopically in all cases.<sup>2</sup> In group 1 the follicles were uniformly small, the epithelium from low cuboidal to cuboidal, and the colloid sparse in amount, staining rather pale pink with eosin (table; fig. 8, left). In groups 2 and 3 the follicles were larger than in group 1, the epithelium flat, and the colloid abundant, staining reddish pink with eosin. The glands were on the whole smaller than in group 1 (fig. 8, right).

The thyroid glands of group 1 (with or without cholesterol feeding) showed evidence of some activity, although the rabbits were killed at the beginning of the summer and had already been partaking of green food. In groups 2 and 3 there was a definite quiescence of the thyroid glands, unquestionably due to the administration of iodine (Webster, Irsigler). The absence of thyroid activity in the latter groups was also indicated by the weights of the animals; they gained from 150 to 500 Gm. (table).

## CHOLESTEROL FINDINGS IN THE BILE AND IN THE LIVER

Whether or not the thyroid gland regulates the cholesterol metabolism is not known. Abelin showed in mice that after the administration of thyroxine the fat content of the organs was reduced before the basal metabolic rate was increased. He expressed the belief that this hormone is specific in regulating the fat requirement. If by the administration of iodine, as in groups 2 and 3, the amount of this hormone is reduced, a study of the excretion of cholesterol becomes important. The bile, being one of the mediums for the excretion of cholesterol (the intestine being the most important excretory organ), was examined. It must be borne in mind that possibilities of error may arise if the bile remains for any period of time in the gallbladder, as concentration may occur.

The normal cholesterol content of the rabbit's bile was found to be between 55 and 75 mg. per hundred cubic centimeters, mainly in the form of free cholesterol (Rothschild's normal values were from 40 to 50 mg. per hundred cubic centimeters). As not all the rabbits had sufficient bile for examination, only a limited number could be examined (table).

For group 1 the cholesterol content was 40 mg. per hundred cubic centimeters; for group 2 b (in the animal in which the highest blood cholesterol was found—2,770 mg. per hundred cubic centimeters—and in which the most marked aortic changes were noted) the cholesterol

2. The animals were killed within one or two weeks in the middle of June. They had already had grass added to their diet for about one month.

content of the bile was 15 mg. per hundred cubic centimeters; for group 3 *a* it varied from 91 to 157 mg.; for group 3 *b* it was 102 mg.

Rothschild found that after twenty-eight days of cholesterol feeding in rabbits the amount of cholesterol in the bile doubled (90 mg. per hundred cubic centimeters). After one hundred and fifty days it was trebled (160 mg.). The cholesterol in the blood was correspondingly increased, from 130 to 364 mg. per hundred cubic centimeters, respectively. Rothschild concluded that the bile cholesterol was proportionate to the blood cholesterol. This is not universally accepted.

Unfortunately the insufficient number of cases does not allow definite conclusions, and as Harley and Barratt have shown, the cholesterol excreted in the bile of herbivorous animals is for the most part resorbed. It is suggestive that in the cholesterol-fed rabbit the liver fails to accommodate quickly enough, and the excretion of cholesterol through the bile is insufficient. A further demonstration of this fact was seen in the determination of the cholesterol content of the liver (table).

Normally, the rabbit's liver contains from 0.11 to 0.13 Gm. of cholesterol (Wacker and Hueck found 0.13 Gm. to be normal). In the cholesterol-fed animals the liver contained from 0.25 to 0.63 Gm. of cholesterol (similar values were obtained by Wacker and Hueck, also Ellis and Gardner). When iodine was fed from the beginning, the amounts of total cholesterol varied from 0.82 to 1.58 Gm. When iodine was added later, the amounts were from 0.89 to 1.54 Gm.

The addition of iodine caused a marked retention of cholesterol not only in the blood but also in the liver. Whatever the action of the iodine was in the cases studied, the end-result seemed to be directed against the excretory parts of the liver.

There is yet to be explained the increase of cholesterol ester in the animals of groups 2 *b* and 3 *b* when the iodine was added later in the course of the cholesterol feeding.

Webster showed that when rabbits with large active thyroid glands are fed massive doses of iodine (7.5 Gm.) they lose a great deal of heat and rapidly die. When smaller doses of iodine are fed (1.5 Gm.) to animals with small active glands, the amount of heat produced is less and the animals survive. He interpreted this loss of heat as an indication of the amount of thyroxine liberated, the latter being proportional to the available iodine. Sections of the thyroid gland taken at frequent intervals during administration of iodine only showed a complete regression of these glands to the colloid type (Webster, Irsigler).

As has been described, the thyroid glands of the animals not fed iodine were slightly to moderately active. When iodine was administered to the cholesterol-fed animals, a regression of the glands probably took place with liberation of thyroxine, as shown by Webster. The increased secretion of this hormone was necessarily followed by an

*Observations in Representative Animals from Each Group*

Rabbit Sex	Type of Feeding	Weight at Start, Gm.	Weight at End, Gm.	Cholesterol in Serum, Mg. per 100 Cc.						Aver. Per- cent- age of Ester	Total Choles- terol in Liver, Gm. per 100 Gc.	Changes in Thyroid Gland	Comment					
				At Start			At End											
				Free	Ester	.....	Free	Ester	.....									
A M	Regular diet only	1.850	....	27	48	...	.....	.....	....	64	55	0.11	Low cuboidal epithelium, little colloid	None				
B M	Regular diet only	1.850	....	22	56	...	.....	.....	....	72	70	0.13	Low cuboidal epithelium, little colloid	None				
83 F	Regular diet + cholesterol in oil	2.200	2,650	23	43.6	96	390	144.4	65.4.6	74	40	0.63	Low cuboidal epithelium, little colloid, follicles small	Slight amount of fat deposited in thoracic por- tion and about issuing vessels				
87 F	Regular diet + cholesterol + potassium iodide	1.950	2,400	30	45	458	2,027	366	2,081	79	..	1.58	Epithelium flat, much colloid in large follicles	Moderate to marked amount of fat in tho- racic portion and slight deposit in abdominal portion				
29 F	Regular diet + cholesterol + Jodotropon	2.075	2,150	11.4	24.85	590	2,070	590	2,070	77	157	0.8	Same as above	Same as above				
26 M	Regular diet + cholesterol, then potassium iodide added	2.350	2,850	34	49	568	2,202	568	2,202	Before Iodine 74	'15	1.54	Same as above	Most marked diffuse deposit in thoracic, and to less extent, in abdominal, portion				
27 F	Regular diet + cholesterol, then Jodotropon added	2.130	2,600	29	42	358	1,064	358	1,064	Before Iodine 74 After Iodine 84	102	0.89	Same as above	Same as above				

increased metabolism which also influenced the liver. As the latter excretes only free cholesterol through the bile (Bürger, Thannhauser), there was a corresponding proportional increase of cholesterol ester in the blood. (The foregoing mechanism is offered only as a suggestion in view of the fact that the entire cholesterol metabolism of the rabbit under these experimental conditions is greatly disturbed.)

After a time when the thyroid gland was inactivated by the iodine, the normal relationship of free cholesterol to cholesterol ester was reestablished. The decrease in the general metabolic activity resulting from the inactivity of the thyroid gland may account for the relatively decreased excretion of cholesterol and for its storage in the body.

Kohno, Kunde, Leupold, Macciota (cited by Bürger), Abelin, and Turner and Khayat have found a decrease in the cholesterol content of the blood after feeding thyroxine, and Rohrschneider and Artoni found an increase after thyroidectomy (without disturbing the parathyroids). Besuglow and Tutkewitsch, by feeding thyroxine to dogs, found that the bile secretion was diminished as well as its cholesterol content, while following thyroidectomy the bile secretion was increased as well as its cholesterol content.

#### COMMENT

The action of iodine in the prevention or the acceleration of deposits of lipoid in the rabbit's aorta as seen from experimental studies is not direct but through the thyroid gland (Turner and Khayat). This activity is probably dependent on increased liberation of the thyroid hormone (Webster).

The doses of iodine given to the animals in this study were large physiologic ones (cf. Webster and Irsigler). That they were not toxic was noted by the inactivity of the thyroid gland, by the absence of microscopic degenerative changes in the aorta or other organs and by the increase in body weight.

The doses of iodine given by Liebig (in the form of an iodized fatty acid ester and by Turner (in the form of potassium iodide) were from 150 to 322 mg. of iodine per day respectively for an animal weighing 2 Kg. If it is assumed that the average human being weighs about 70 Kg., this would mean a daily consumption of from 5 to 10 Gm. of iodine (from 7 to 13 Gm. of potassium iodide). As Liebig has shown, if the iodine is excluded from the rabbit's diet for a short period (two or three weeks), the protective action of the iodine is to a great extent lost. The results given by Seel and Creuzberg, who used small doses of iodine (2 mg. of potassium iodide), could not be substantiated by me.

Granted that the cholesterol content of human blood rarely reaches the heights obtained in the rabbit, the point that is driven at is that the action of iodine is not a direct one but through its activation of the thyroid gland, and that the large physiologic doses cause regression of a gland that may be slightly active, with a resulting increase of the lipoid deposit in the aorta. Massive doses of iodine given over a longer period may not only be toxic directly (Liebig, Pfeiffer) but may stimulate the thyroid gland to activity (Raab, Pfeiffer). Indeed, it is only by the latter method that the prevention of atherosclerosis in the rabbit is brought about. If large physiologic doses are administered, an increased rather than a decreased deposit may take place.

From a clinical standpoint, the cholesterol content of the blood is decreased in exophthalmic goiter (Epstein and Lande, Kohno, Bing and Heckscher, cited by Friedland). Hurxthal found that usually under 100 mg. of cholesterol per hundred cubic centimeters of blood indicated a very toxic state, while over 200 mg. indicated a state varying from mildly toxic to normal (in hyperthyroidism). On administering compound solution of iodine, the blood cholesterol rose as the toxicity decreased. In some instances an increase of the blood cholesterol was found, but then the phosphorus fraction was also increased, speaking for destruction of nerve tissue (Omelyanovich-Pavlenko).

Atherosclerosis in persons with hyperthyroidism is uncommon (also Aschoff). On the contrary, in athyreosis, myxedema and cretinism, in which the blood cholesterol is increased (Luden, Heckscher, cited by Friedland), the incidence of atherosclerosis is higher (Heine, Bourneville). Wegelin and also Hellwig state that a definite myxedematous state is not necessary but that a mild to moderate hypofunction of the thyroid gland will suffice.

#### SUMMARY AND CONCLUSIONS

Large physiologic doses of iodine in the form of inorganic potassium iodide or organic Jodtropon produced a marked increase of the cholesterol in the blood and liver and a corresponding decrease of the cholesterol in the bile in cholesterol-fed rabbits. The hypercholesterolemia thus produced was far beyond that of rabbits which had had cholesterol alone added to their diet.

When iodine was added later in the course of cholesterol feeding, the cholesterol esters of the blood increased for six weeks. It is suggested that in the rabbits in which the cholesterol metabolism was so markedly disturbed the action of the iodine on a slightly active gland caused a regression of the gland and in doing so liberated an increased amount of the thyroid hormone. The latter stimulated the general metabolism as well as the liver. As the liver excretes only free cholesterol through the bile, there was a corresponding proportional

increase of the cholesterol esters of the blood. The aortas of the latter animals presented the most marked lipoid deposit, stressing the importance of the cholesterol esters in experimental atherosclerosis.

A comparison of the thyroid glands of the animals fed iodine with those of the animals not fed iodine revealed a resting state of the colloid in the former and a slightly active state in the latter.

As the action of iodine in the prevention of deposits of lipoid in the aorta of the cholesterol-fed rabbit is dependent on increased thyroid activity, its employment in man should be guarded.

#### BIBLIOGRAPHY

- Abelin, I.: Klin. Wchnschr. **10**:2205, 1931.  
Anitschkow, N.: Beitr. z. path. Anat. u. z. allg. Path. **51**:379, 1913; Experimental Arteriosclerosis in Animals, in Cowdry, E. V.: Arteriosclerosis: A Survey of the Problem, New York, The Macmillan Company, 1933, p. 271.  
Artoni, C.: Arch. di sc. biol. **5**:22, 1923-1924.  
Aschoff, L.: Verhandl. d. deutsch. path. Gesellsch. **10**:166, 1906; Beitr. z. path. Anat. u. z. allg. Path. **47**:1, 1910; Beihefte z. med. Klin. **1**:1, 1930.  
Besuglow, V. P., and Tutkewitsch, L. M.: Ztschr. f. d. ges. exper. Med. **87**:52, 1933.  
Bourneville, D.: Arch. de neurol. **16**:38, 1903.  
Bürger, M.: Ergebn. d. inn. Med. u. Kinderh. **34**:583, 1928.  
Ellis, W., and Gardner, J. A.: Proc. Roy. Soc., London **84**:461, 1911-1912.  
Freund, H., and König, W.: Arch. f. exper. Path. u. Pharmakol. **33**:317, 1928.  
Friedland, I. B.: Ztschr. f. d. ges. exper. Med. **87**:683, 1933.  
Guggenheimer, H., and Fisher, I. L.: Ztschr. f. d. ges. exper. Med. **54**:114, 1927.  
Gumprecht, F.: Verhandl. d. Kong. f. inn. Med. **19**:260, 1901.  
Harley, V., and Barratt, W.: J. Physiol. **29**:341, 1903.  
Heine, J.: Beitr. z. path. Anat. u. z. allg. Path. **72**:590, 1924.  
Hellwig, A., and Neuschlosz, S. M.: Klin. Wchnschr. **1**:1988, 1922.  
Hildebrandt, F.: Arch. f. exper. Path. u. Pharmakol. **96**:292, 1923.  
Hurxthal, L. M.: Arch. Int. Med. **52**:86, 1933.  
Irsigler, F. H.: Beitr. z. path. Anat. u. z. allg. Path. **85**:221, 1930.  
Janeway, F. C.: Boston M. & S. J. **174**:925, 1916.  
Liebig, H.: Arch. f. exper. Path. u. Pharmakol. **159**:265, 1931.  
Mancke, R.: Arch. f. exper. Path. u. Pharmakol. **149**:56, 1930.  
Müller, O., and Inada, R.: Virchows Arch. f. path. Anat. **283**:282, 1932.  
Murata, M., and Kataoka, S.: Tr. Jap. Path. Soc. **8**:221, 1918.  
Omelyanovich-Pavlenko, M.: Klin. Med. **9**:761, 1931.  
Pfeiffer, G.: Endokrinologie **13**:40, 1933.  
Potain, P. C. E.: Clinique médicale de la Charité, Leçons et mémoires, Paris, G. Masson, 1894.  
Raab, W.: Hormone und Stoffwechsel, in Boas, F.; Neuberg, C., and Rippel, A.: Naturwissenschaft und Landwirtschaft, Munich, Dr. F. P. Datterer & Cie, 1926, pt. 10.  
Rohrschneider, W.: Virchows Arch. f. path. Anat. **256**:139, 1925.  
Romberg, E.: Lehrbuch der Krankheiten des Herzens, Stuttgart, Ferdinand Enke, 1921.  
Rothschild, M. A.: Beitr. z. path. Anat. u. z. allg. Path. **60**:39, 1915.

- Schönheimer, R.: Virchows Arch. f. path. Anat. **251**:732, 1924; **249**:1, 1924;  
Ztschr. f. physiol. Chem. **160**:61, 1926; **177**:143, 1928.
- Seel, H., and Creuzberg, S.: Arch. f. exper. Path. u. Pharmakol. **161**:674, 1931.
- Shapiro, S.: J. Exper. Med. **45**:595, 1927.
- Stockman, R., and Charteris, F.: Brit. M. J. **2**:1520, 1901.
- Thannhauser, S. J.: Deutsches Arch. f. klin. Med. **141**:290, 1922-1923; Lehrbuch  
des Stoffwechsels und der Stoffwechselkrankheiten, Munich, J. F. Bergmann,  
1929.
- Thoussig, R.: Wien. med. Wchnschr. **52**:1399, 1902.
- Turner, K. B.: J. Exper. Med. **58**:115, 1933.  
—and Khayat, G. B.: J. Exper. Med. **58**:125, 1933.
- Wacker, L., and Hueck, W.: Arch. f. exper. Path. u. Pharmakol. **71**:373, 1912-  
1913.
- Webster, B.: Studies in the Experimental Production of Simple Goiter, Interna-  
tional Goiter Conference, Bern, 1933, p. 1.
- Wegelin, C., in Henke, F., and Lubarsch, O.: Handbuch der speziellen patholo-  
gischen Anatomie und Histologie, Berlin, Julius Springer, 1926, vol. 8, p. 1.

# ARGENTAFFINOMAS OF THE GASTRO-INTESTINAL TRACT, BENIGN AND MALIGNANT

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BOSTON

Oberndorfer<sup>1</sup> described a peculiar type of tumor of the gastro-intestinal tract and called it "carcinoid" because of its resemblance to carcinoma in histology but not in behavior. Gosset and Masson<sup>2</sup> showed that tumors of this type contain cytoplasmic granules which reduce silver ammonium oxide. Their work further demonstrated that these granules are present in the Kultschitzky cells of the intestinal mucosa. These had been described by Kulitschitzky<sup>3</sup> as specialized epithelial cells in the crypts of Lieberkühn. The cells have been shown to occur occasionally in the stomach and to become fairly numerous in the duodenum while in the jejunum and ileum they are moderately abundant. They are most frequent in the appendix, but they can be demonstrated in small numbers in the colon (Maximow and Bloom<sup>4</sup>). Their function is not known, though Masson<sup>5</sup> suggested the possibility of endocrine secretion. Masson<sup>6</sup> and others have stated that these tumors are closely related to nerve fibers. By the use of a trichrome stain, Masson<sup>6a</sup> followed the budding of the Kulitschitzky cells from the crypts of Lieberkühn and their subsequent migration into the nerve sheath of the underlying hyperplastic nerves or neuromas. In this location, the Kulitschitzky cells proliferate and form tumors. Because of the presence of the specific silver granules in the cytoplasm of these neoplastic cells, the term "argentaffinoma" seems more descriptive than "carcinoid."

The earlier writers regarded all these tumors as benign. However, isolated reports of metastasizing argentaffinomas have appeared in the literature, and recently the more frequent occurrence of malignancy in this type of tumor has been recognized. Raiford<sup>7</sup> described six malignant argentaffinomas and collected twenty-eight additional instances

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1. Oberndorfer, S.: Frankfurt. Ztschr. f. Path. **1**:426, 1907.

2. Gosset, A., and Masson, P.: Presse méd. **22**:237, 1914.

3. Kulitschitzky, N.: Arch. f. mikr. Anat. **49**:7, 1897.

4. Maximow, A., and Bloom, W.: Textbook of Histology, Philadelphia, W. B. Saunders Company, 1931.

5. Masson, P.: Compt. rend. Acad. d. sc. **158**:59, 1914.

6. Masson, P.: (a) Am. J. Path. **4**:181, 1928; (b) ibid. **6**:499, 1930.

7. Raiford, T. S.: Am. J. Cancer **18**:803, 1933.

from the literature. The same series was discussed later by Lewis and Geschickter<sup>8</sup> with the addition of two more cases. Besides these Masson and Martin<sup>9</sup> described a case of malignant argentaffinoma of the stomach. Christopher<sup>10</sup> presented an additional case with the primary tumor in the ileum. The total number of malignant argentaffinomas recorded, then, is thirty-eight.

It seems that comparison of the benign and malignant argentaffinomas may provide useful data in relation to their type of growth. Is the complexity of the stroma maintained outside the primary growth? Are all the elements represented in all the metastatic nodules or does the representation vary? To what extent is the character of the stroma dependent on the adjacent tissue? It would seem that those elements which follow the tumor beyond its original site and are seen in the metastases must be stimulated by the tumor cells.

#### MATERIAL

The material available for this study consisted of thirty-one argentaffin tumors of the gastro-intestinal tract obtained from twenty-one patients during the past seven years at the Peter Bent Brigham Hospital. The cases are described briefly in the table. These tumors occurred among 10,668 specimens of all types, a total incidence of 0.2 per cent. One had produced remote metastases, one was locally invasive, two invaded the musculature without penetration of the serosa, while the remaining twenty-seven tumors were entirely benign. One tumor was located in the stomach and was found incidentally at autopsy. A single benign argentaffinoma was located in the jejunum. The ileum was the site of sixteen tumors. This group included a locally invasive specimen as well as a tumor producing metastases to the liver and mesentery. Thirteen were seen in the appendix, two of which invaded the musculature, while all the others were confined to the mucosa and submucosa. None of these showed extension beyond the appendical serosa. There were no instances of argentaffin tumors of the colon in this series.

The entire series was studied microscopically in routine sections. Three of the tumors were chosen as representative of the various stages in argentaffinomas. The ones selected were a rather large benign tumor of the stomach, a locally invasive argentaffinoma of the ileum and an obstructive tumor of the ileum with metastases to the mesentery and liver. Sections from these were stained by many methods.

8. Lewis, D., and Geschickter, C. F.: Arch. Surg. **28**:16, 1934.

9. Masson, P., and Martin, J. F.: Bull. Assoc. franç. p. l'étude du cancer **17**:139, 1928.

10. Christopher, F.: Surg., Gynec. & Obst. **58**:903, 1934.

*Data in Thirty-One Cases of Argentaffinoma of the Gastro-Intestinal Tract*

Patient	Sex	Age	Symptoms, Etc.	Location	Metastases
A	F	66	Reported in full. Case 1.....	Stomach	None
B	M	59	Death following resection of sigmoid for adenocarcinoma; argentaffin tumor incidental finding at autopsy	Jejunum	None
C	F	66	Acute purpura with secondary anemia (cause undetermined); tumor incidental finding at autopsy	Ileum	None
D	F	71	Generalized arteriosclerosis; myocardial infarction; tumors incidental finding at autopsy	Ileum (9)	None
E	F	41	Rheumatic heart disease with aortic and mitral stenosis and insufficiency; tumor incidental finding at autopsy	Ileum	None
F	F	64	Death following cholecystectomy for empyema of gallbladder; acute suppurative pancreatitis; multiple abscesses of liver; tumor incidental finding	Ileum	None
G	M	36	Tuberculoma of brain; bilateral tuberculosis of suprarenals; tumor incidental finding	Ileum	None
H	M	57	Reported in full. Case 2.....	Ileum (3)	One locally invasive; two benign
I	M	64	Reported in full. Case 3.....	Ileum	Mesentery, liver
J	F	17	Acute abdominal pain of twelve hours' duration, starting about umbilicus; spasm and tenderness in right lower quadrant	Appendix	None
K	F	20	Irregular menses, backache, ventral suspension of uterus; appendectomy	Appendix	None
L	F	21	Vague pain and tenderness constantly in right lower abdominal quadrant for seven years; acute exacerbation without nausea during that period; examination showed slight tenderness in right lower quadrant, maximal at McBurney's point; appendectomy	Appendix	None
M	F	17	Intermittent attacks of pain in right lower abdominal quadrant for two years; appendectomy	Appendix	None
N	F	41	Recurrent attacks of nausea, vomiting and constipation for six weeks; progressive enlargement of abdomen; supravaginal hysterectomy for leiomyomas of the uterus; incidental appendectomy	Appendix	None
O	M	28	Epigastric pain for two years, relieved by food and alkalis; excision of pylorus and adjacent duodenum for multiple peptic ulcers; incidental appendectomy	Appendix	None
P	F	41	Dull pain in lower part of abdomen for six days with constipation; bilateral salpingectomy for acute and chronic salpingitis; incidental appendectomy	Appendix	None
Q	F	44	Pain in lower part of back and dragging sensation in lower part of abdomen; uterine fixation and repair of pelvic floor; incidental appendectomy	Appendix	None
R	F	15	Acute abdominal pain, nausea and vomiting for twenty hours; diarrhea; no muscle spasm; temperature 99 F.; pulse 100; respirations 20; appendectomy	Appendix	Invasion of muscularis; no extra-appendical extensions; well two years later
S	F	35	Recurrent attacks of pain in right lower abdominal quadrant, accompanied by nausea; appendectomy; no attacks after operation	Appendix	None
T	F	20	Pain in right lower abdominal quadrant for five days; temperature 100 F.; tenderness, without muscle spasm, in right lower quadrant; operation showed acute salpingitis and periappendicitis; appendectomy	Appendix	None
U	F	25	Repeated attacks of pain in right lower quadrant for several years; severe pain maximal at McBurney's point for three days; appendectomy	Appendix	Invasion of muscularis; no extra-appendical extensions

## PREPARATION OF MATERIAL

*Fixation.*—Tissue fixed in solutions of formaldehyde or Kaiserling's fluid was available in each case. Zenker's fluid also was used in cases 1 and 2, while in case 1 material had been fixed also in Bouin's fluid.<sup>10a</sup> In order to make use of stains requiring fixation in Zenker's fluid in the blocks from case 3, they were carried through ammonia by Davidoff's<sup>11</sup> method, mordanted in Zenker's fluid, and embedded. Sections from other paraffin blocks were mordanted on the slide.

*Staining Methods.*—All stains were applied to each tumor. For general study and nuclear detail hematoxylin-eosin and eosin-methylene blue were used. The stains for the intercellular substances were Mallory's aniline blue-acid fuchsin-phosphomolybdic acid-orange G, Van Gieson's hematoxylin-trinitrophenol-acid fuchsin, and Mallory's phosphotungstic acid-hematoxylin stain. Weigert's resorcinol-fuchsin stain was used to demonstrate elastic fibers. This was followed by various nuclear stains, usually basic fuchsin. Masson's<sup>6a</sup> method for the argentaffin granules ("silvering the sections") was used in all instances. Reticulum was studied by the Cajal-Maresch technic, followed by Van Gieson's trinitrophenol-acid fuchsin staining. This silver method stained the argentaffin granules. An attempt was made to secure silver impregnation of the nerves by Foot's<sup>12</sup> modification of Roger's method for paraffin sections, since this was developed especially to demonstrate the finest nerve fibers. As in the methods used by Masson, no satisfactory impregnation of the nerves was secured. However, the argentaffin granules were stained very intensely, so that the cytoplasm of the cells was filled with reduced silver. This was an undesirable result from the standpoint of histologic study but was an advantage in securing low power photomicrographs (fig. 7). The intense reaction was considered to depend on the pretreatment with ammoniated alcohol followed by 40 per cent silver nitrate.

## CASE 1: BENIGN ARGENTAFFIN TUMOR OF STOMACH

*Clinical Data.*—M. H., a white woman 66 years old, was admitted to the hospital in a stuporous condition. She was found to have rheumatic heart disease. She had an excellent appetite. The bowels were regular, but the stools were nearly always loose. No melena was noted at any time. Death followed an extensive facial erysipelas. Autopsy showed the cardiac lesion to be aortic stenosis. A benign argentaffin tumor of the stomach was an incidental finding.

*Gross Description.*—The only lesion with which this investigation was concerned was the tumor of the stomach. The stomach was small and contracted. Externally no puckering of the serosa or infiltration of the gastrohepatic omentum was seen. There were no metastases demonstrable in any organ. The serosa was smooth, pale gray and glistening in all parts. The stomach was opened by a single incision along the greater curvature. It contained a small amount of partially digested food. No ulceration of the mucosa was noted at any point. On the lesser curvature, 4 cm. below the cardiac orifice, there was a tumor measuring 13 mm. in diameter. It was located in the submucosa, for it was covered by a

10a. The formula for Bouin's fluid (Masson's modification) is: 40 per cent formaldehyde, 10 parts; water, 30 parts; glacial acetic acid, 2 parts, and trinitrophenol to saturation.

11. Davidoff, L. M.: Am. J. Path. 4:493, 1928.

12. Foot, N. C.: Am. J. Path. 8:769, 1932.

thin uninterrupted layer of mucosa which was slightly tense about its edges, while it could be moved freely over the tunica muscularis. There was no contraction of the gastric wall about the tumor. Neither here nor elsewhere in the stomach was there any change in the thickness of the tunica muscularis. A single cross-section of the tumor was made in the median plane. It was covered by an uninterrupted, thin layer of mucosa which was fused partially with the tumor. The cut section was yellowish gray and was firm in consistency. It was not entirely homogeneous, occasional tiny gray areas being seen scattered through the yellow tissue. No areas of necrosis or of hemorrhage were found. The entire freedom of the muscularis from invasion or distortion by the tumor was striking. The serosa was entirely normal.

*Microscopic Description.*—The surface was covered at all points by a thin but intact layer of mucosa. The glands were normal in size and arrangement. Kultschitzky cells were found in small numbers near the bases of the glands while the parietal and chief cells were present in their usual proportions. Spreading out from the lower portion of the mucosa was a tumor. While its connection with the mucosa could be traced, the bulk of the nodule occupied the submucosa. It was entirely discrete though no definite capsule was seen about the tumor. The type cell was a small one with an oval nucleus and a scanty cytoplasm. The cytoplasm was filled with very numerous argentaffin granules. Each cell was demarcated clearly in most portions from those adjacent to it, and often in the stained section there were spaces between the cells without reticulum or other stromal fibers. In other areas the tumor cells were arranged compactly. There were no mitoses, and giant cells were not seen. The cells in all parts of the tumor were strikingly uniform in size and shape. The stroma divided the tumor cells into rather large masses. A few very delicate reticular fibers passed into the masses but at the centers these were not seen. The larger groups of stromal fibers were complex in structure. There were large numbers of smooth muscle fibers in compact groups while a few of these extended into the finer ramifications of the stroma. In close association with the muscle fibers there was an unusual degree of hyperplasia of the elastica. By study of comparable sections stained with Weigert's resorcinol-fuchsin and Mallory's aniline blue connective tissue stain, it was noticed that the areas in which smooth muscle fibers were most numerous were the locations in which the increase in elastica was greatest. Collagen fibers were present in moderate numbers. These were associated with an abundant reticular network which was confined mostly to the larger masses of stromal fibers, passing occasionally into the larger clumps of tumor cells. No nerve fibers could be impregnated by the Foot-Rogers<sup>12</sup> technic. A careful study of sections stained with Mallory's aniline blue connective tissue stain and Mallory's phosphotungstic acid-hematoxylin stain left some doubt as to the presence of nerve hyperplasia. At any rate it was not so prominent as the increase in elastica or in smooth muscle. On the whole, the tumor contained few blood vessels. These had a single layer of elastica for the internal elastic membrane. There was a definite hyperplasia of the elastica in the adventitia, the fibers extending for a short distance into the tumor as an integral part of the stroma. No areas of necrosis were found. The tunica muscularis was uniform in thickness beneath the tumor. There was no evidence of tumor invasion or distortion of this structure. The serosa showed no change.

CASE 2: ARGENTAFFIN TUMOR OF THE ILEUM WITH INVASION OF  
THE MESENTERY BUT WITHOUT REMOTE METASTASES;  
MULTIPLE BENIGN ARGENTAFFINOMAS

*Clinical Data.*—M. O., a white man 57 years of age, entered the hospital for treatment of a cerebral tumor. He had not had any symptoms which could be referred to lesions of the gastro-intestinal tract. A large frontal meningioma was removed. On the thirtieth day after operation the patient died of pulmonary embolism following thrombosis of the left iliac vein. The argentaffin tumors were incidental findings at autopsy.

*Gross Description.*—The lesion with which this investigation was concerned was an argentaffin tumor located 2 cm. above the ileocecal valve. A firm, grayish-white mass was found in this position along the mesenteric attachment. This extended about half way around the circumference of the ileum and protruded from the serosal surface as a slightly elevated, puckered nodule. The ileum proximal to the tumor mass was very slightly dilated while distally it was normal in diameter. The mesenteric fat was adherent to the tumor and on section was seen to contain firm tissue of fibrous consistency in large amounts. The dense connective tissue extended along the adjacent mesentery for 3 cm. At the limit of extension of the tumor into the mesentery there was a firm mass 0.3 cm. in diameter. It was demarcated clearly from the adjacent fat, which was puckered and contracted.

After fixation, the ileum was opened along the free margin. It could be seen then that the tumor arose from the mesenteric border of the intestine and projected into the lumen so as to narrow it but slightly. The diameter of the ileum was little larger above the tumor than below. The lymphoid tissue of Peyer's patches was not altered. The nodule of tumor itself was 10 mm. in greatest longitudinal diameter and 5 mm. in elevation above the surrounding ileal mucosa. The mucosa was intact in all parts. A cut section showed tissue of firm, resilient consistency. No ulcerations of the thin mucosa could be found. The submucosa was widened, but the bulk of the nodule was composed of the hypertrophied and distorted tunica muscularis. The circular layer was very thick, and in a longitudinal section its fasciculi radiated from a puckered, densely fibrous center, being separated from each other by delicate cords of tumor. The neoplasm here as in the mesentery presented a pale yellow, firm surface without any evidence of hemorrhage or necrosis.

Two benign argentaffin tumors were found in the ileum of this subject. One, 4 mm. in diameter, was located 2 meters, and another; 2 mm. in diameter, was seen 1.5 meters, from the ileocecal valve.

*Microscopic Description.*—Over the surface of the tumor there was a thin layer of mucosa. Masses of tumor cells could be seen throughout the mucosa but most often in close relation to the bases of the glands. However, most of the neoplasm was situated in the submucosa with extension into the muscularis and through it to the mesentery. The type cell was a small one with an oval nucleus and scanty granular cytoplasm. Such cells tended to grow in masses, which varied considerably in size. There was very little evidence of cell membrane, the cytoplasm of adjacent cells blending closely. It contained many argentaffin granules. Mitoses were not found after careful examination. There were no tumor giant cells. The type of cell making up the tumor was remarkably uniform in all portions. The stroma of the neoplasm was composed of various elements. Smooth muscle was abundant. This was derived in part from the hypertrophied muscularis

mucosae, but it extended into the smaller ramifications of the stroma, in which place its derivation could not be traced with certainty. Elastic tissue was present as an integral part of the stroma, being most abundant in those locations in which there were groups of smooth muscle fibers, but extending freely into other portions. Collagen fibers were seen in moderate numbers and were associated with a delicate reticulum. There was a moderate degree of hypertrophy of the circular layer of the tunica muscularis. This was invaded by small groups of argentaffin cells with but little destruction of the muscle fasciculi. The tumor tended to grow in the deeper layers beyond the muscularis. In this outer portion, the stroma was composed almost entirely of very dense collagenous tissue. The distortion produced by this part of the tumor was considered the cause for the buckling of the circular layer of muscle. The extensions into the mesentery were similar to the portions of the tumor already described. No demarcation could be made out in the sections between them and the intra-intestinal growth. In the blood vessels no changes in the intima were noted. There was a slight hypertrophy of the elastica in the adventitia. No neoplastic cells were seen within endothelial-lined spaces.

Sections through the mesentery parallel with the long axis of the intestine showed cords of tumor invading the fat. The stroma was made up of very dense collagenous tissue, accounting for the puckering of the fat which was noted grossly. In the tumor were several nerves. Some of them were invaded by the tumor. Elastic tissue fibers were even more numerous than in the primary tumor. Sections of numerous mesenteric lymph nodes showed no metastatic neoplasm. No tumor cells were seen within lymphatics.

#### CASE 3: MALIGNANT ARGENTAFFIN TUMOR OF THE ILEUM WITH METASTASES TO THE MESENTERY AND LIVER

*Clinical Data.*—C. S., a white man 64 years old, entered the hospital complaining of severe constipation, which he had had six weeks. He had been entirely well until two years before admission, at which time he had an attack of constipation associated with nausea and vomiting. This subsided without medical attention after five days. During the intervening two years he had several similar attacks, the last of which occurred six weeks previously. After that, there were intermittent nausea and vomiting, which were accompanied by mild abdominal distention and cramplike pain in the epigastrium and about the umbilicus. The patient lost 17 pounds (7.7 Kg.) in weight during the six weeks. No definite history of melena could be obtained. Physical examination showed an emaciated elderly man in acute pain. The abdomen was slightly distended. Synchronous with the attacks of pain a mass that was tender could be palpated in the left lower quadrant of the abdomen. Between paroxysms of pain, the abdomen was free from masses. On the day following admission a large segment of ileum and its mesentery was resected. Ileocecostomy was performed. The ileum contained a constricting tumor, and there were masses of neoplastic tissue in the mesentery. At that time, tumor nodules could be palpated in the liver. None of these was removed. After operation, the patient did well. He was no longer troubled by distention of the abdomen, and he could have normal bowel movements. He was discharged from the hospital on the thirty-first day after operation. When seen six months later the patient was symptom-free.

*Gross Description.*—The specimen consisted of a portion of the distal ileum measuring 148 cm. in length. About the midpoint of the specimen there was a

constriction roughly dividing it into two equal parts. The constriction was formed by a tumor, which was seen as a firm spherical mass measuring 15 by 15 mm. in its greatest dimensions (fig. 2). A triangular portion of mesentery was attached, which measured 80 mm. in length. At its apex was a mass of tissue which was composed not only of what appeared to be lymph nodes but also of a considerable amount of cicatrized fat tissue into which some tumor had infiltrated. The nodes were gathered in a cluster and together with the regional fat formed a mass which measured 4 by 4 by 3 cm. On section this large firm mass presented along its distal half a fairly well demarcated group of nodules which seemed to be encap-

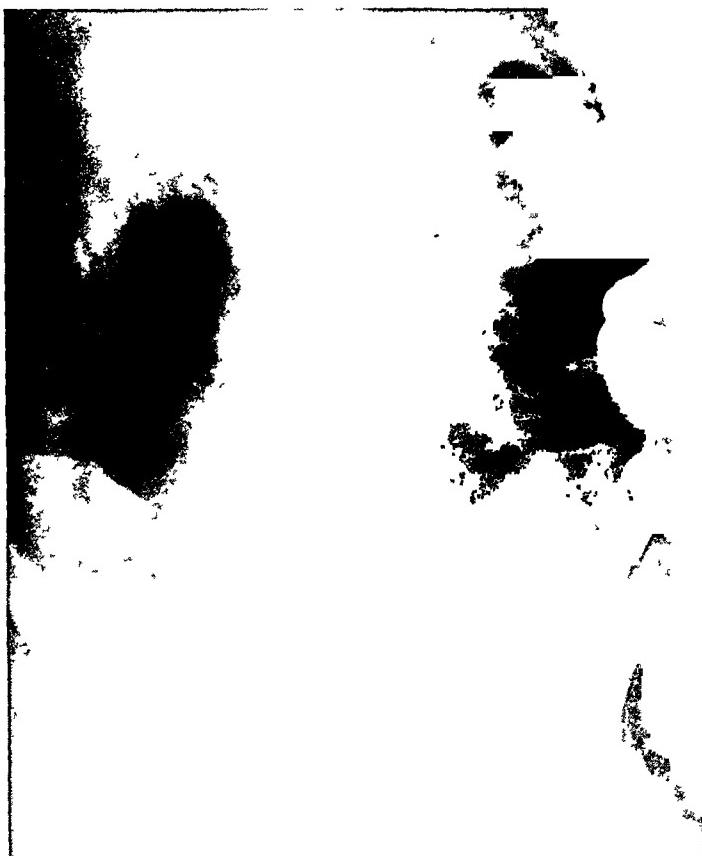


Fig. 1 (case 3).—Complete obstruction of the small intestine caused by a malignant argentaffinoma. The colon contains barium. Note the enormous distention of the loops of small intestine which the slowly progressive nature of the obstructing lesion permits. This type of roentgenogram has been seen in several cases reported in the literature.

sulated by the fibrous tissue. The peculiar lemon yellow color of the neoplasm was striking. These nodules were uniform in texture and number except in occasional areas where there was a brownish-black pigmentation. Along the distal margin of the nodules there was a definite zone of dense connective tissue which demarcated the firm yellow neoplasm from the regional fat comprising the remainder of the tumor in the mesentery. The bowel mucosa throughout its extent was intact. The Peyer's patches, which were few in number, were small and showed



Fig. 2 (case 3).—Photograph of the gross surgical specimen. The lesion appears annular, but the intestine pouches out unevenly above the constriction.



Fig. 3 (case 3).—The intestine has been opened. The lesion encircles only a part of the circumference. Note the dilatation of the intestine above the lesion. No ulceration is evident.

no lesions. Proximal to the constricting lesion the villous folds which encircled the lumen of the bowel were quite prominent, averaging 3 mm. in height a short distance above the obstruction. There was also some hypertrophy of the muscularis, so that this layer measured 1 mm. in thickness. At a distance of 6 cm. above the obstructing lesion the villous folds became somewhat flattened out and lost their prominence. At this point the ileum was 14 cm. in circumference. At the point of obstruction it was 2 cm. in circumference. This narrowing of the lumen was due largely to the presence of a constricting tumor which was situated opposite the attachment of the mesentery. The lumen was approximately 2 mm.

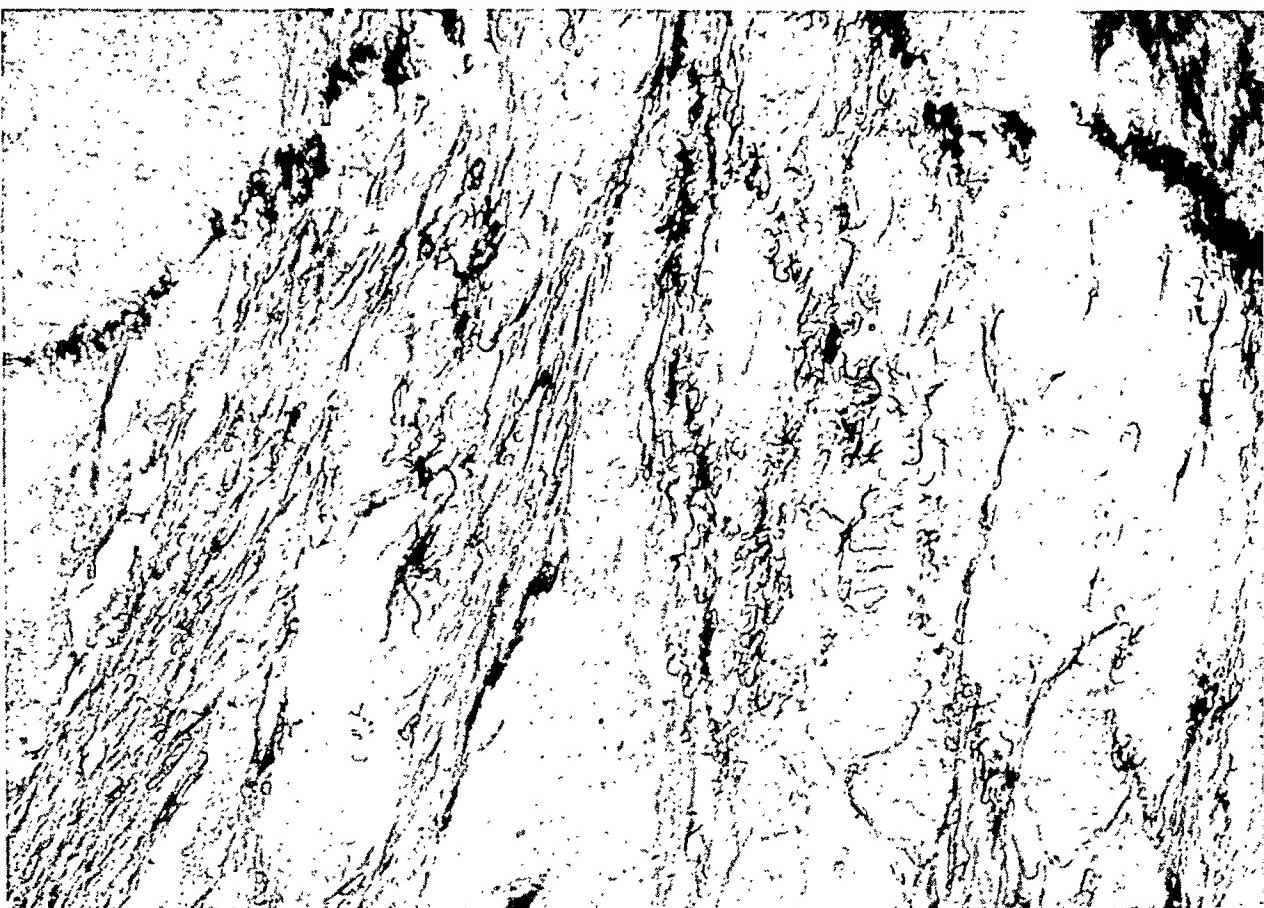


Fig. 4 (case 3).—Weigert's resorcinol-fuchsin stain counterstained with eosin;  $\times 210$ . This shows the abundance of elastica in the primary tumor.

in diameter but was partially closed by the folds of mucosa so that a probe could be passed only with difficulty. The tumor nodule involved no more than half the circumference of the ileum (fig. 3). It measured 18 by 12 by 12 mm. Beneath the mucosal surface and the tunica muscularis there was a zone of tumor which averaged 4 mm. in thickness. The tunica muscularis was drawn sharply in and angulated, forming a horseshoe-shaped coat of muscle with the convexity toward the lumen and the open end of the angulation at the serosal surface. The muscularis seemed to be interrupted in a few areas by tumor, but there was extension of the tumor through the tunica muscularis so as to replace and to invade the serosa. The fatty tissue was puckered at the mesentery as if invaded by neoplasm.

*Microscopic Description.*—Blocks from the primary tumor and its mesenteric metastases were studied. In addition, serial blocks of mesentery from the primary tumor to the metastases were examined.

The primary tumor had the characteristic structure of an argentaffinoma. At each margin of the tumor there was ileal mucosa of average thickness. Its glands were uniform and contained only the usual number of argentaffin cells, these seldom exceeding three to a gland. The muscularis mucosae was normal. No hyperplasia



Fig. 5 (case 3).—Photomicrograph showing the growth of the tumor in the extramuscular layers with buckling of the tunica muscularis;  $\times 9$ .

of smooth muscle, nerves or elastic tissue was noted. In the center of the tumor it was seen that the overlying mucosa was preserved very completely. From the bases of the deeper glands small groups of tumor cells extended downward into the submucosa. These were more or less independent of each other. They soon coalesced and in the submucosa formed a mass of neoplastic tissue. Here the cells were arranged in large masses into which a few reticular and elastic tissue fibers penetrated. The individual cells were small with oval nuclei, which contained delicately arranged chromatin material and which usually showed a single

nucleolus, staining especially well with the metachromatic silver technics. The cytoplasm occupied a relatively small proportion of the total cell volume. At the edges of the cell masses and in the actively advancing part of the tumor the cytoplasm was filled with fine argentaffin granules indistinguishable from those of the Kultschitzky cells of the intestine. The centers of the cell masses did not preserve these granules so well, though a few could be found in most cases. No mitoses were seen in any of the tumor cells. An alveolar arrangement was recognized frequently in which a row of cells surrounded a small lumen. The nuclei were adjacent to the lumen, the argentaffin granules being at the base of the cell just



Fig. 6 (case 3).—Weigert's resorcinol-fuchsin stain lightly counterstained with basic fuchsin;  $\times 210$ . This shows the proliferation of elastic fibers in the adventitia of a blood vessel in the mesentery with extension of these fibers into the tumor.

as in normal Kultschitzky cells. There were instances of reduplication in layers so as to form the masses of indistinctly demarcated cells farther from the lumen.

At the center, the muscularis mucosae blended with the stroma but could be recognized at the edges even within the confines of the tumor. The stroma was of great complexity and appeared to be derived from the muscularis mucosae, the submucosa and the tunica muscularis. In this tumor specific staining technics showed that all the elements of the submucosa, as well as of the muscular layers above and below it, participated in the formation of the stroma of the argentaffinoma, and not the neuromuscular complex alone. There was judged to be a slight

hyperplasia of nerve fibers, which could not be impregnated by the Foot-Rogers technic. The smooth muscle was more abundant and passed throughout the dense cord of tissue which separated the neoplastic masses. A delicate argyrophilic reticulum was found in all parts of the tumor stroma. Of special interest was the increase in elastic tissue fibers. These at times formed dense limiting membranes about cell masses and at others were matted together with the smooth muscle or extended as single strands along the stroma. In the thick bands of connective tissue which separated the groups of tumor cells, collagenous fibers were abundant.

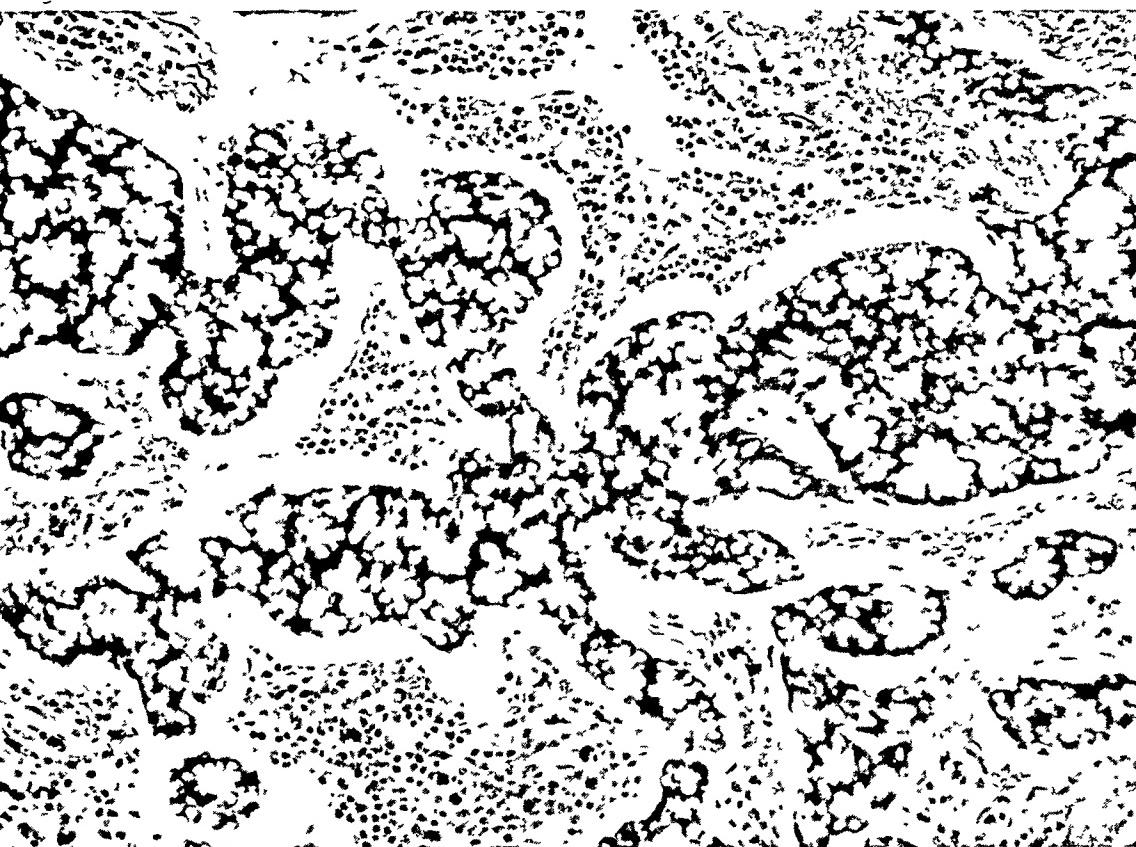


Fig. 7 (case 3).—Mesenteric metastasis; Foot-Rogers method;  $\times 325$ . This shows small groups of cells invading a lymph node. The argentaffin granules are intense black. No nerves are impregnated. The alveolar arrangement is more pronounced here than in most parts of the tumor.

Only a small portion of the constricting tumor was found in the submucosa. The muscularis was hypertrophied enormously about a central core made up of tumor with a dense fibrous stroma which caused scarring and distortion, so that a section taken longitudinally showed the muscle fasciculi spread out like the ribs of a fan (fig. 5). Small groups of argentaffin tumor cells were seen scattered through the tunica muscularis. Coarse elastic tissue fibers accompanied the fasciculi, and a dense group of fibers of this type followed the outer border of the circular layer somewhat like a limiting membrane. Beyond this point, larger groups of tumor cells were found, and the same complex stroma intervened. On

the whole, the neoplasm was not very vascular, and no tumor cells were seen within endothelial-lined spaces.

The tissue between the primary growth and the metastasis was examined by serial blocks 3 mm. thick. Strands of tumor cells followed blood vessels freely through the fat. These blood vessels showed a marked hyperplasia of the elastic tissue of the adventitia with extension of these fibers into the tumor as though the tumor were stimulating the formation of elastic tissue fibers for its stroma from this source. The intima was thickened, and reduplication of the internal

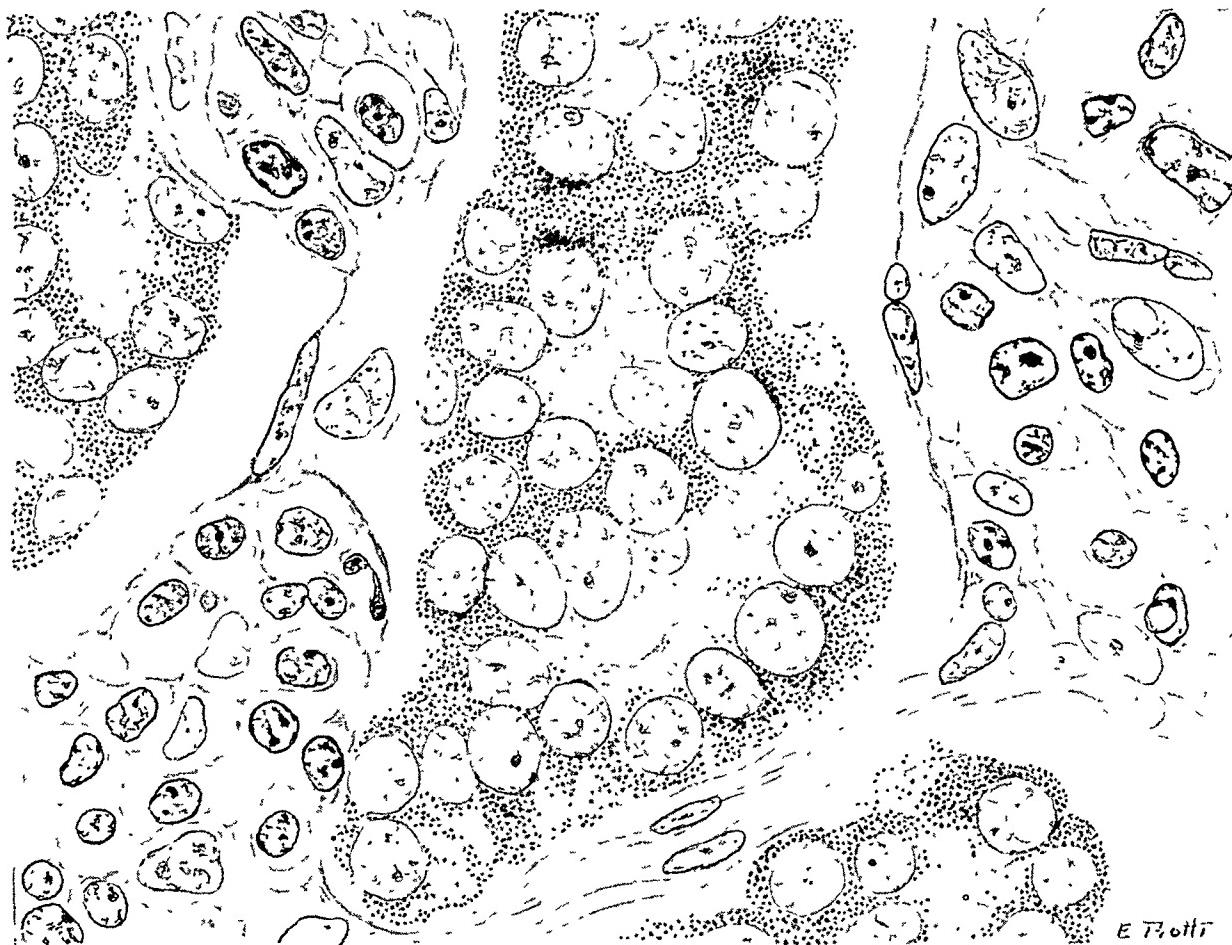


Fig. 8 (case 3).—Camera lucida drawing,  $\times 1,300$ , showing a portion of a mesenteric lymph node infiltrated by metastatic argentaffinoma. The staining is by Masson's method for "silvering the sections," counterstained with alkaline methylene blue. In the center and at each side of the drawing there are clusters of tumor cells separated from each other by lymphoid tissue. The cytoplasm of each tumor cell is filled with specific argentaffin granules.

elastic membrane was seen in some vessels. There was also some hypertrophy of the smooth muscle of the media. No tumor cells were seen within lymphatics.

The metastasis was situated, not in lymph nodes primarily, but just outside. The nodes were invaded by small groups of actively growing tumor cells. The metastasis was composed chiefly of groups of argentaffin cells with cytoplasms poorly demarcated from one another. Among these groups very little reticulum

or other stromal fibers extended. The smaller groups showed a greater tendency to alveolar formation. The argentaffin granules stained very intensely. The stroma was made up of very dense bands of collagenous tissue in which smooth muscle was identified in small amounts. Thin, delicate elastic tissue fibers were seen especially in the dense outer covering of the tumor. They closely paralleled the distribution of smooth muscle. No association of nerves with the stroma of the mesenteric cords or with that of the metastasis was apparent.

Unfortunately, no material from the metastasis in the liver was secured. The presence of a malignant tumor of the small intestine in connection with nodules of tumor in the liver would be presumptive evidence of their identity.

#### COMMENT

A series of thirty-one argentaffin tumors of the gastro-intestinal tract has been presented with a detailed description of three of them. Each of the three is considered representative of a different type in relation to malignancy. The large tumor of the stomach was entirely benign and has been chosen to represent this group because the total volume of argentaffin tissue was approximately the same as that of the intra-intestinal portion of the other tumors. The second case was one of local invasion; the third, of definite malignancy with production of distant metastases.

Each of these tumors consists of two parts, and each part has its special interest. The argentaffin cells are clearly the neoplastic ones. Their origin from the Kultschitzky cells of the intestine cannot be questioned. However, the germ layer from which the Kultschitzky cells are derived is by no means certain. Masson<sup>6b</sup> and Maximow and Bloom<sup>4</sup> stated that they are entodermal in origin. This is suggested by their intimate connection with the entodermal cells of the mucosal glands, in which location they can be recognized by the characteristic silver reduction of their granules as early as the fourth month of fetal life, much sooner than the granules of the cells of Paneth can be identified. Schack<sup>13</sup> confirmed this view. He stated that in the embryo the argentaffin cells appear first in the mucosal glands and that their presence elsewhere is the result of subsequent migration. Kull,<sup>14</sup> however, expressed the belief that the Kultschitzky cells arise from the mesoderm and migrate early in fetal life to lie in their close association with the entodermal elements of the mucosa. Raiford,<sup>7</sup> on the other hand, considered that an ectodermal origin could be postulated because of the association with nerves as described by Masson and because argyrophilic properties are a common characteristic of ectodermal cells while those of the entoderm do not show such an affinity. However, the silver impregnation of many mesodermal structures is recognized widely. Entodermal cells frequently contain the Golgi apparatus, which can be

13. Schack, L.: Beitr. z. path. Anat. u. z. allg. Path. **90**:441, 1932-1933.

14. Kull, H.: Ztschr. f. mikro-anat. Forsch. **2**:163, 1925.

demonstrated by silver impregnation. In later communications Masson<sup>6</sup> reiterated his belief that these cells are of entodermal origin. If argentaffinomas are inseparable from nerves, the nerves concerned are entodermal too, in his opinion.

The stroma of argentaffinomas is remarkable for its complexity. The presence of a hyperplasia of the nerves has been emphasized by Masson.<sup>6a</sup> He considered the tumor to begin as a neuroma, with subsequent migration of the argentaffin cells into the nerve sheath. In this location these cells undergo neoplasia. However, other writers have been unable to demonstrate a constant hyperplasia of the nerves, and the migration within the nerve sheath has not been seen by several investigators, including Raiford,<sup>7</sup> who looked with special care for it. Hyperplasia of the nerves has not been demonstrated beyond the intestinal serosa by any observer. Case 2 in this series showed invasion of nerve trunks in the mesentery.

Like all epithelial tumors argentaffinomas present in their stroma a variable number of collagenous fibers. These fibers vary in number, but more particularly in collagen production, in different locations. In the tumors studied the stroma contained a moderate number of such fibers in the mucosa, submucosa and tunica muscularis. However, in the masses of tumor in cases 2 and 3 which formed at the serosa there was very extensive collagen formation. This same dense collagenous tissue surrounded the mesenteric metastasis in case 3, while the cords of tumor extending between the primary growths and this point were accompanied by very delicate fibers of this type. In all tumors of this type argyrophilic reticular fibers are numerous, especially in association with the less dense collagen. While the reticulum extends into the masses of tumor cells farther than the other fibers, there are masses of argentaffin cells into which no stromal element can be seen to penetrate.

The presence of smooth muscle fibers in the stroma of the intra-intestinal section has been recognized by many observers. These fibers appear to be derived in part from the muscularis mucosae and in part from the tunica muscularis. The formation of smooth muscle is stimulated at the sites in which it is normally present. In the present material one could never be sure of its derivation from the media of blood vessels. Furthermore, smooth muscle as a stromal element has not been found in the mesenteric extension or in the mesenteric metastasis of the malignant ones. It occurs in groups, occasionally ending in isolated muscle cells in the smaller stromal masses.

No special studies have been found in the literature on the elastic tissue element in argentaffinomas. Accordingly, these fibers have been studied in detail in all locations. Elastica is found to be very abundant in both benign and malignant tumors of this type. At times the fibers are found in clusters and at others they are scattered through the

collagenous tissue passing into the finer radices of the stroma. Usually it is associated in location with smooth muscle. When large masses of elastica are found, comparable sections stained with Mallory's aniline blue connective tissue stain demonstrate smooth muscle in the same location. This is an association pointed out in the case of nerve fibers. In case 3 the elastica was found in the stroma of the tumor cords in the mesentery and in that of the metastasis. It thus does not show the restriction to the intra-intestinal portions of argentaffinomas seen with smooth muscle and nerve fibers. In several places, but most clearly in case 1 and in the cords of tumor in the mesentery in case 3, the origin of the elastic tissue fibers from the adventitia of small and medium-sized blood vessels has been demonstrated. In such areas, there is a hyperplasia of the elastica in the adventitia with extension of the fibers among the masses of tumor cells, the number of fibers becoming progressively less the greater the distance from the vessel (fig. 6). Whether all the elastic tissue has such an origin cannot be stated with certainty. It is considered more probable that much of that seen in the intra-intestinal tumors is derived from the elastica normally present in the submucosa. Thus, these tumors are shown to have the property, apparently rare among tumors, of stimulating the production of elastic tissue fibers. In exceptional cases of many types of tumor elastic fibers are found in abundance. These are largely, if not wholly, the elastica of the tissue invaded by the neoplasm. They are not stimulated by the desmoplastic properties of the tumor. Such findings are not constant for any particular type of neoplasm. In all the instances of argentaffinoma studied, an abundance of elastic tissue fibers has been seen. The proliferation of the elastica of the adventitia of blood vessels is a striking feature and illustrates the ability of these tumors to stimulate new formation of elastic tissue.

As a final note on the stroma of argentaffinomas it should be pointed out that the amount of any one special element of the stroma varies from place to place and from tumor to tumor. Collagen fibers and reticulum are always present. The other elements are present to some degree in almost all instances, but any one may predominate in a given place. The type of fiber seems to depend in large part on the variety of normal tissue adjacent to the tumor.

Besides these characteristics of the argentaffinomas and their stroma, several features of the malignant type should be pointed out. The percentage of malignancy increases with advancing years. It might be supposed that these tumors have been present for very long periods before symptoms develop. For example, in a patient in Raiford's<sup>7</sup> series a malignant argentaffinoma was found at operation in a patient 16 years of age. It was located in the appendix and metastases were present in the mesentery. The only symptoms were those of chronic

appendicitis. It is interesting to speculate on the possibility that without operation this patient would have lived for many years before the symptoms usually associated with a malignant tumor in this location developed, by which time distant metastases might have been present. In the two cases showing malignancy in the present series the patients are both at the upper limit of the age group, 57 and 64 years. Such tumors may have been developing for very long periods of time.

The cases of malignant argentaffinoma recorded in the literature (including the present ones) were distributed at the different levels of the gastro-intestinal tract as follows:

Stomach .....	2
Duodenum .....	0
Jejunum .....	2
Ileum .....	19
Small intestine (exact location not specified).....	8
Appendix .....	7
Colon .....	2

Raiford<sup>7</sup> pointed out that benign argentaffinoma is much more common in the appendix than elsewhere with decreasing frequency at progressively more distant levels, so that the percentage of malignancy is lowest in the appendix and increases in proportion to the distance from that point. In his series, those of the stomach and colon were all malignant. However, the argentaffin tumors in these locations are so few that conclusions would be less definitive in respect to them than in respect to those in the appendix. The only argentaffinoma of the stomach in the series reported here was benign. None were found in the colon.

As judged from the two cases of malignant argentaffinoma studied, the mechanism of intestinal obstruction is different in these tumors from that of the usual adenocarcinoma of the intestine. The latter tumor produces narrowing of the lumen by growing entirely around the intestine in an annular manner with further constriction resulting from the contraction of the connective tissue in the stroma. The malignant argentaffinoma grows only a part of the way around the intestinal lumen (fig. 3) even though the external appearance of the unopened specimen, as in case 3, suggests an annular constriction (fig. 2). Low power microscopic examination (fig. 5) shows that the tumor growing from the mucosa into the submucosa and infiltrating the hypertrophied muscularis extends into the serosal layer. Here it forms a mass of tumor which has a dense stroma composed largely of collagenous fibers. The progressive contraction at this point causes a buckling of the overlying intestinal layers. There is formed in consequence a rounded mass, which projects into the lumen. The uninvolved intestine adjacent to the mass is puckered and distorted by this process with the result that the lumen is almost completely occluded. That this is the fundamental mechanism is suggested by the fact that in those tumors which

do not extend into the muscularis no distortion of the intestinal contour results. Furthermore, this same finding is present in cases in which intestinal narrowing is seen. In case 3, in which obstruction was almost complete, the extramuscular mass of tumor was larger than in case 2, in which the intestinal lumen was still functionally patent.

It should be pointed out that argentaffinomas show little or no ulceration. This is in contrast to the adenocarcinomas of the same location, which ulcerate extensively.

The mesenteric metastases, as seen in case 3 and as described repeatedly in the literature, occur characteristically outside lymph nodes, while these structures are invaded by smaller groups of actively proliferating tumor cells. Between the primary growth and the mesenteric metastases, as in case 3 and in the mesentery adjacent to the intra-intestinal growth in case 2, cords of tumor can be seen which infiltrate the fat freely. Many of these follow blood vessels and derive their stroma in part from the adventitia of such vessels. No evidence of lymphatic invasion was seen in this series.

Metastases are present in many cases beyond the limits of the mesentery. The nodules palpated in the liver at operation in case 3 were in all probability of such a nature. A tabulation of the metastases seen in cases reported in the literature (including the present cases) is as follows:

Mesentery (lymph nodes).....	31
Liver .....	16
Cecum .....	1
Ileum .....	1
Peritoneum .....	1
Omentum .....	2
Pelvis .....	1
Lungs .....	1
Pleura .....	1

b

Lewis and Geschickter<sup>8</sup> advanced the theory that argentaffinomas are paragangliomas of the gastro-intestinal tract, differing only in location from paragangliomas of the suprarenal gland or carotid body. There are certain considerations which make such a view at least improbable. The Kultschitzky cells are entodermal or possibly mesodermal derivatives with little evidence pointing to an ectodermal origin. As seen in this study, given fields in benign and malignant argentaffinomas are histologically very similar, the differentiation being only on the behavior of the tumor. Lewis and Geschickter's statement on paragangliomas of the suprarenal gland is: "In the malignant forms giant, atypical ganglion and large spindle cells are found. Malignant paragangliomas are remarkable because of the size and variety of the giant cells. Eisenberg and Wallerstein have said that the greatest imaginable irregularity in size and shape of the cells and nuclei is the most striking histologic feature." Such a picture is not seen in malignant argentaffinomas, regularity in cell type being notable. Paragangliomas do not possess

the characteristic stroma described in argentaffin tumors. Masson<sup>15</sup> considered the argentaffinomas as paragangliomas of a hypothetic entodermal system and hence not in any way related to ectodermal tumors. The evidence points strongly to a place apart for the argentaffinomas, a grouping with tumors elsewhere in the body appearing impossible at the present time.

The clinical manifestations of argentaffin tumors are of some interest. The benign type except for those in the appendix are absolutely asymptomatic. The remarkable freedom of these tumors from ulceration and their small size contribute to this. In the series reported here no instance of intussusception resulting from an argentaffinoma was encountered, and the literature examined yields no report of such a case. It is beyond the scope of this paper to discuss the production of an internal secretion by the Kultschitzky cells or by the tumors arising from them. However, in the cases studied, the tumor was associated with disease of the endocrine glands only once, in that instance with tuberculosis of the suprarenal glands. Tumors made up of cells sufficiently differentiated to contain specific granules might be expected to function if they arise from endocrine tissue. No evidence of such disturbance could be found.

When the appendix is the site of an argentaffinoma, whether benign or malignant, symptoms are present which are not encountered in association with these tumors elsewhere. The patient has attacks of dull pain in the right lower abdominal quadrant which may be associated with acute manifestations, such as sharper pain, nausea and vomiting without fever or leukocytosis. Masson<sup>16</sup> described this syndrome as "neurogenic appendicitis." He pointed out that small neuromas are found in a high percentage of cases of so-called chronic appendicitis. This finding was confirmed by Hosoi.<sup>17</sup> Furthermore, Masson<sup>18</sup> stated the belief that these neuromas form an integral part of the development of the argentaffinomas. It is difficult to see why a similar process at a different level would be so entirely asymptomatic. However, in this series, the tumors of patients J, L, M, R, S and U (table) belonged to this group. One might raise the question whether patient N was not benefited by the removal of the appendix instead of by the "major" part of the operation, consisting in the removal of lesions on the basis of which it is difficult to explain attacks of nausea and vomiting.

The malignant argentaffinomas produce symptoms very late. Ninety-two and five-tenths per cent were not discovered until lymph node or even more remote metastases were present. The clinical manifestations are those of chronic intestinal obstruction. As pointed out by Raiford,<sup>7</sup>

15. Masson, P.: Am. J. Path. **6**:217, 1930.

16. Masson, P.: Compt. rend. Acad. d. sc. **173**:262, 1921.

17. Hosoi, K.: Arch. Path. **16**:500, 1933.

diarrhea without melena is a frequent finding in the earlier stages. Indeed, one seldom sees the anemia so often associated with other types of malignant growth in the same location. The absence of ulceration in malignant as well as in benign argentaffinomas accounts largely for this finding. The intestinal obstruction is insidious, yet progressive. It allows time for tremendous dilatation of the proximal loops of bowel. In case 3 roentgenograms showed such a change (fig. 1). This was seen in several cases reported in the literature, notably that of Gáspár.<sup>18</sup>

The prognosis of malignant argentaffin tumors from a statistical review of the recorded cases appears to be decidedly unfavorable. However, the nature of the changes has been recognized in several cases only at postmortem examination, the location or even the presence of a malignant condition not being suspected during life. The tumors have a remarkably slow rate of growth. If found before remote metastases are present, the chance of recurrence will be much less than that of the usual adenocarcinoma in a similar location.

The treatment of malignant argentaffinomas must be surgical. A radical resection is indicated clearly in all cases without generalized metastases. When masses are found in the mesentery, these require excision of that sector with its attached intestine, often a meter or more. Such resection offers palliation for long periods of time even with metastases palpable in the liver, as in case 3. Because of the tendency of the argentaffinomas to invade the mesentery in cords very early, a constricting lesion of the gastro-intestinal tract which is recognized as a malignant argentaffinoma at the time of operation by its yellow color and by the peculiar method of growth described in this paper requires the resection of a sector of mesentery even in the absence of gross invasion. The percentage of cures in patients treated in this way will in all probability be very high.

#### SUMMARY

A series of thirty-one argentaffinomas of the gastro-intestinal tract is described. Three cases are presented in detail as representative of the various types of these tumors.

The stroma of the argentaffinoma consists of collagen fibers, reticulum, nerve fibers, smooth muscle cells and elastica. A study of the elastic tissue shows it to be abundant in both the benign and the malignant type of argentaffinoma. The metastatic tumor has a stroma composed of collagen, reticulum and elastic fibers. The elastica is derived in part from the adventitia of the small and medium-sized blood vessels in the tumor. The argentaffinoma has the property of stimulating the production of elastic tissue, a characteristic rare among tumors.

18. Gáspár, I.: Am. J. Path. 6:515, 1930.

Intestinal obstruction results from the growth of the tumor in the serosal layer with consequent buckling and distortion of the tunica muscularis. In this series, at least, complete annular tumor growth did not occur.

Material is presented to show that the argentaffinoma cannot be regarded as a paraganglioma of the sympathetic nervous system.

Clinically, the argentaffinoma in any location except the appendix is characterized by its extremely asymptomatic course—throughout life if the tumor is benign; if malignant, until very late in the natural history of the tumor. With location in the appendix, the symptoms of acute or chronic appendicitis are simulated frequently. This is true whether the tumor is benign or malignant.

The extremely slow growth of the tumor of the malignant type encourages radical resection even in the presence of metastases. The tendency of the tumor to grow in cords in the mesentery renders it advisable to resect a segment of mesentery even in the absence of gross invasion.

Because of the asymptomatic course of the malignant tumors until very late, few have been subjected to early operation. Of those that did, very few have recurred.

# ALLERGIC INFLAMMATION OF THE LUNGS

## THE PATHOGENESIS OF LOBAR PNEUMONIA

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In previous articles I<sup>1</sup> have reported studies on the reaction of the lungs to bacteria and other substances in normal animals. The present contribution deals with the response of the lungs in animals that have been rendered allergic to a foreign protein.

### MATERIAL AND METHOD

The experiments were conducted on full-grown rabbits fed on oats, hay, and green vegetables.

Five cubic centimeters of horse serum (diphtheria antitoxin) was injected intraperitoneally every sixth day over a period of forty-eight days, the amount of serum totaling 40 cc. On the sixth day after the last injection 1 cc. of serum was injected into the lung via the trachea. The technic of the operation has been described in a former publication.<sup>2</sup> The animals were killed from one hour to four weeks after the intratracheal ("shocking") injection.<sup>3</sup> The lungs fixed *in situ* were cut at different planes through the entire thickness, and sections stained with different dyes were studied.

### OBSERVATIONS

*Macroscopic Appearance.*—The gross appearance of the lungs depended on the interval between the intratracheal injection and the death of the animal. When the rabbit was killed from eight to ten hours after the "shocking" injection, the surfaces of the lungs appeared normal. But when the thoracic cavity was opened from twelve to fifteen hours after the intratracheal injection, one lung (the right in the majority of cases) showed a smooth firm area of discoloration in the dependent portion of the lung and along the spinal column, resembling somewhat the paravertebral pneumonia occasionally seen in children. Of seventy-five rabbits used in the experiments, 60 per cent showed the following:

In some instances both lungs were of the same size; in others, either one whole lung or the lower lobe was somewhat shrunken, smooth and firm, while the

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This investigation was aided by a grant from the Emanuel Libman Fellowship Fund.

1. Fried, B. M.: Arch. Path. **3**:46, 1927; **6**:1008, 1928; **10**:213, 1930; **12**:689, 1931; **17**:76, 1934.

2. Fried, B. M., and Whitaker, L. R.: Arch. Int. Med. **40**:726, 1927.

3. The word "shocking" is not appropriate; it is used for want of a better term.

other lung was normally wrinkled, crepitant and collapsible. When placed in water the firm lung sank, pulling down its apparently normal fellow. A condition in which the size of one lung was reduced is shown in figure 1 *A* and *B*. From this illustration it may be seen that the lower lobe of the left lung is contracted, its base showing two areas of discoloration. It is noteworthy that the pulmonary surface was covered with fibrin, and that a sizable sterile fibrinous exudate, made up of

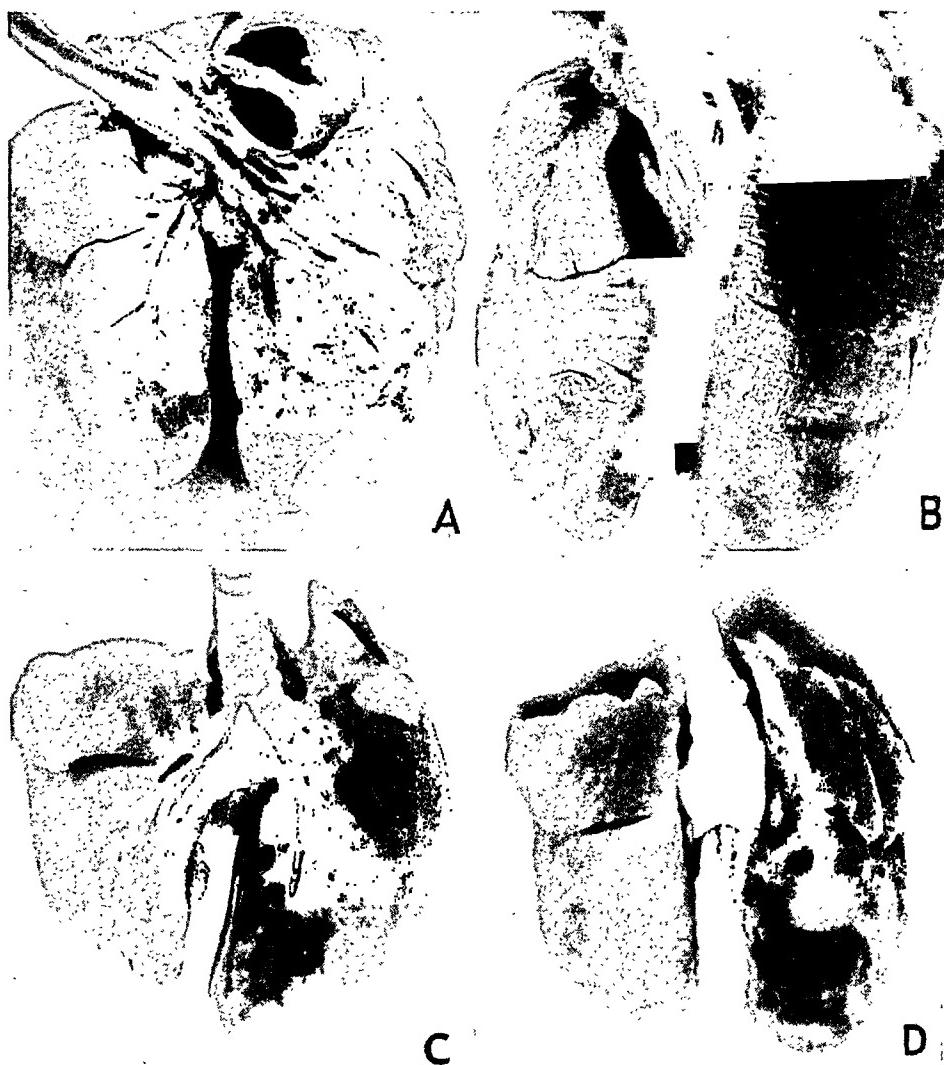


Fig. 1.—Pneumonic lungs. *A*, cut surface of the left lung; *B*, pleural surface of the same lung covered with fibrin; *C*, cut surface of the right lung; *D*, pleural surface of the same lung showing areas of discoloration (infarctions).

macrophages and granulocytes, was present in the pleural cavity. The cut surface of these lungs revealed "hepatization" of the left lung with a large hemorrhage—an infarct—in the lower segment of the organ (fig. 1 *A* and *B*). The picture resembled that of acute lobar pneumonia as seen in man.

Figure 1 *C* and *D* is illustrative of a group of animals in which the right lung was involved. In these cases both lungs were usually equal in size, but occasionally

the affected one was slightly larger (fig. 1 C and D). Nearly all these animals showed wide subpleural hemorrhages (infarctions). The cut surface of the lungs of the rabbits killed two and four days respectively after the injection of the "shocking" dose is demonstrated in figure 2. In these animals the left lung showed uniform consolidation of nearly half of the lower lobe, while the right lung was normal. The affected areas in both of the lungs were vaguely demarcated, and their cut surfaces were finely granular. The color of the lungs varied; the lesion of two days' duration was dark red, while the four day lesion was grayish white, showing evidence of a healing process.

*Microscopic Examination.*—Microscopically, the response of the parenchyma to the "shocking" dose was conspicuous within one hour. The lesion was that of an exudation of granulocytes and serum into the alveoli leading to their distention. The septal capillaries showed marked congestion and a large number of polymorphonuclear leukocytes mingling with erythrocytes. The reaction rapidly increased in intensity, and after three or four hours granulocytes flooded the lung,

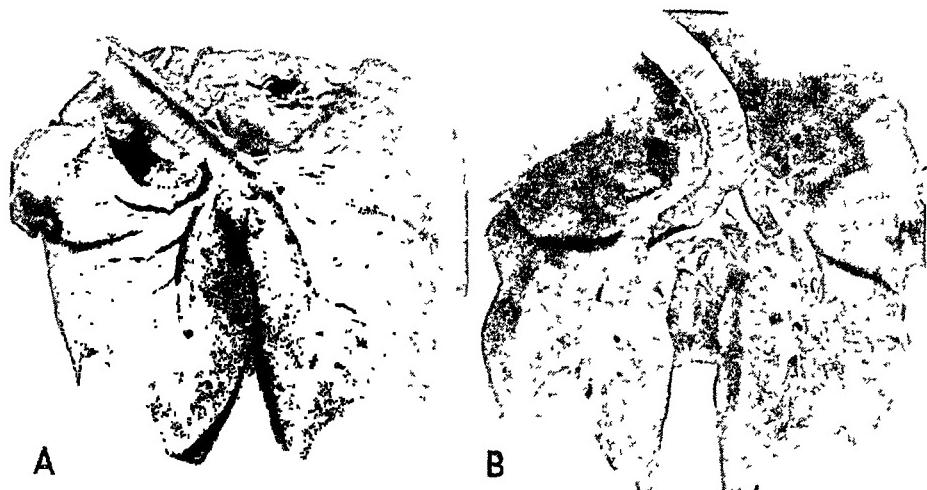


Fig. 2.—Cut surface of the pneumonic lungs. *A*, lungs removed from an animal killed two days after the "shocking" injection of serum; *B*, lungs removed four days after the "shocking" injection. In both cases the left lung is affected.

resulting in a tremendous dilatation of the air sacs. A thickening of the septums resulted from the massive accumulation of leukocytes and the swelling of the alveolar "epithelial" cells. Because of the abundance of the cellular exudate the normal markings of the lungs were obliterated in many areas. Insular atelectasis also occurred. That the cells were brought to the lungs by the circulating blood was apparent from their overwhelming numbers in the capillaries and larger blood vessels. The small vessels were plugged with granulocytes, squeezing out the erythrocytes.

A rapid disintegration of leukocytes was found in the lung of animals that were killed about twenty-four hours after the injection of the "shocking" dose. Wide areas of these cellular débris could be seen enmeshed in a coarse network of fibrin. At this time the lungs of a large number of animals contained an exudate composed of two varieties of mononuclear cells: macrophages (alveolar "epithelial" cells) detached *en masse* from the septums, and large eosinophilic cells apparently brought with the circulation (fig. 3*A*). In a few instances the latter

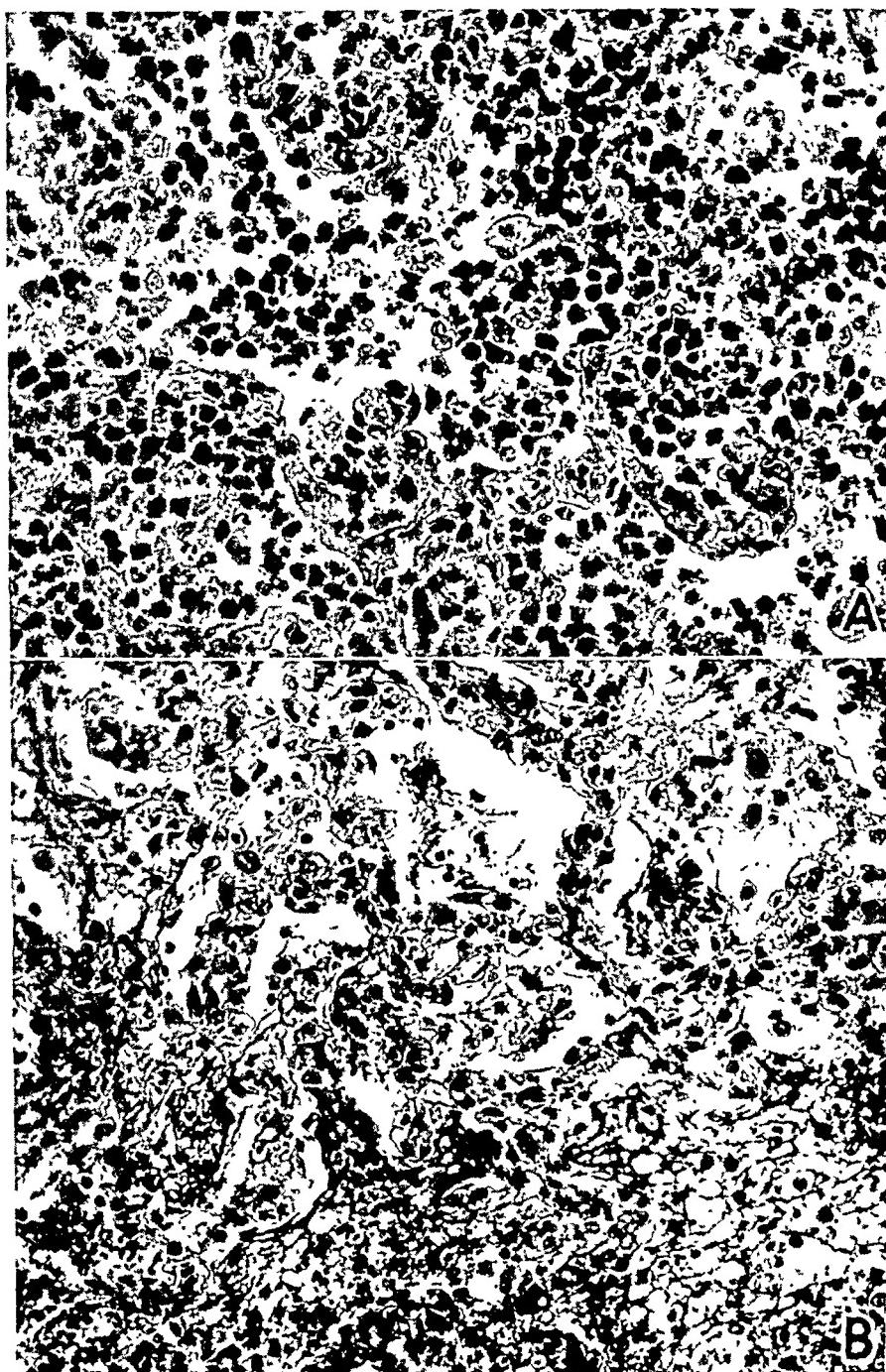


Fig. 3.—*A*, exudate in the pneumonic lung; *B*, meshwork of fibrin containing entangled mononuclear cells and disintegrated granulocytes.

cells dominated the picture. The alveolar macrophages were occasionally in mitoses and proliferated along the septums forming in places continuous files.

Fibrin began to appear in from twenty to thirty hours after the injection of the "shocking" dose, reaching its acme in approximately forty-eight hours (fig. 3 B).

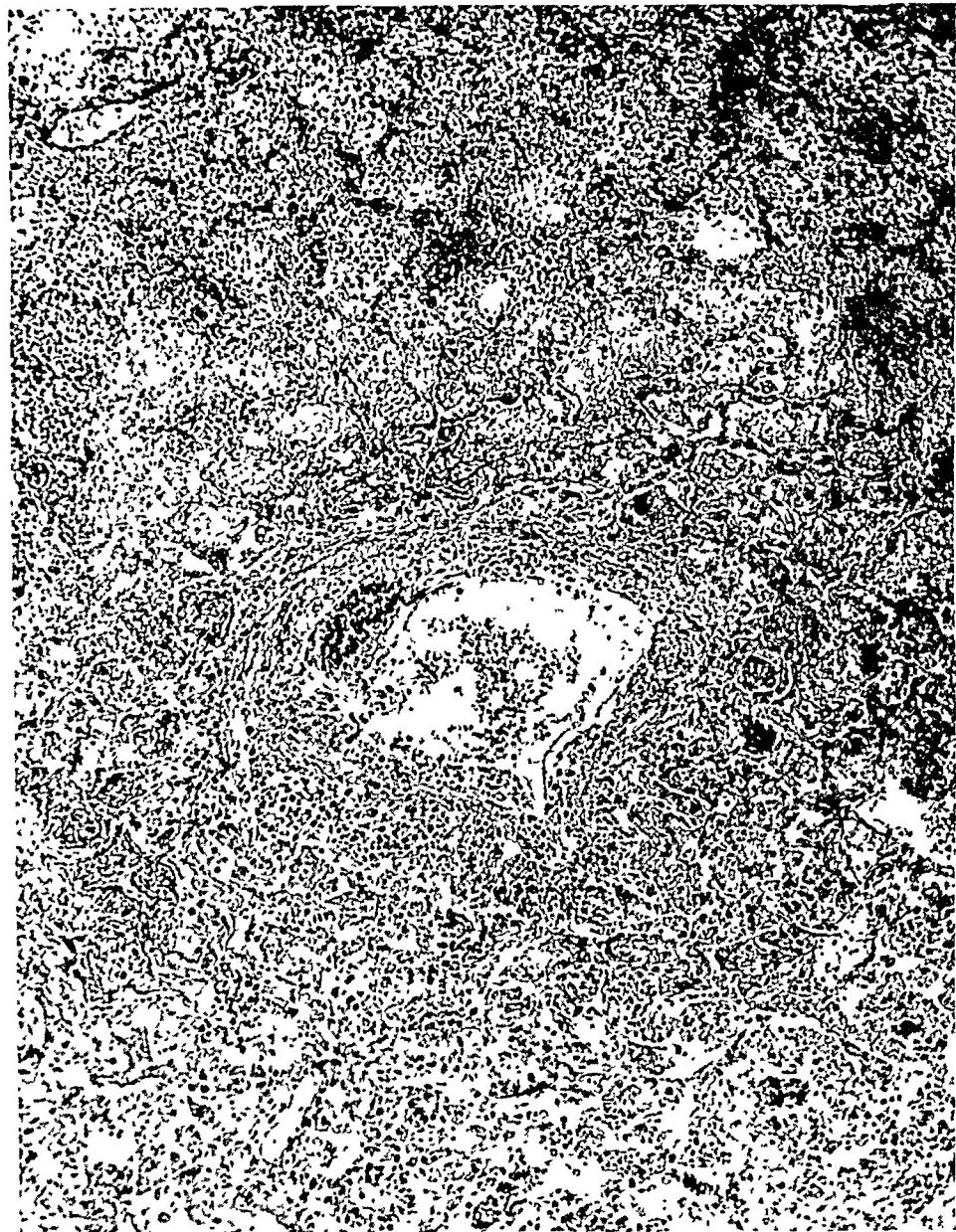


Fig. 4.—Condition of a blood vessel found in the midst of the pneumonic parenchyma. The animal was killed twenty-four hours after the intratracheal ("shocking") injection of horse serum.

At that stage markings in the lungs were largely obliterated. There was a coarse meshwork of fibrin in which "corpses" of granulocytes were entangled, while the far more numerous mononuclear cells appeared to be rather normal.

The duration of the reaction paralleled the amount of pulmonary tissue involved. For example, in the rabbits (about 20 per cent) in which the inflammatory process

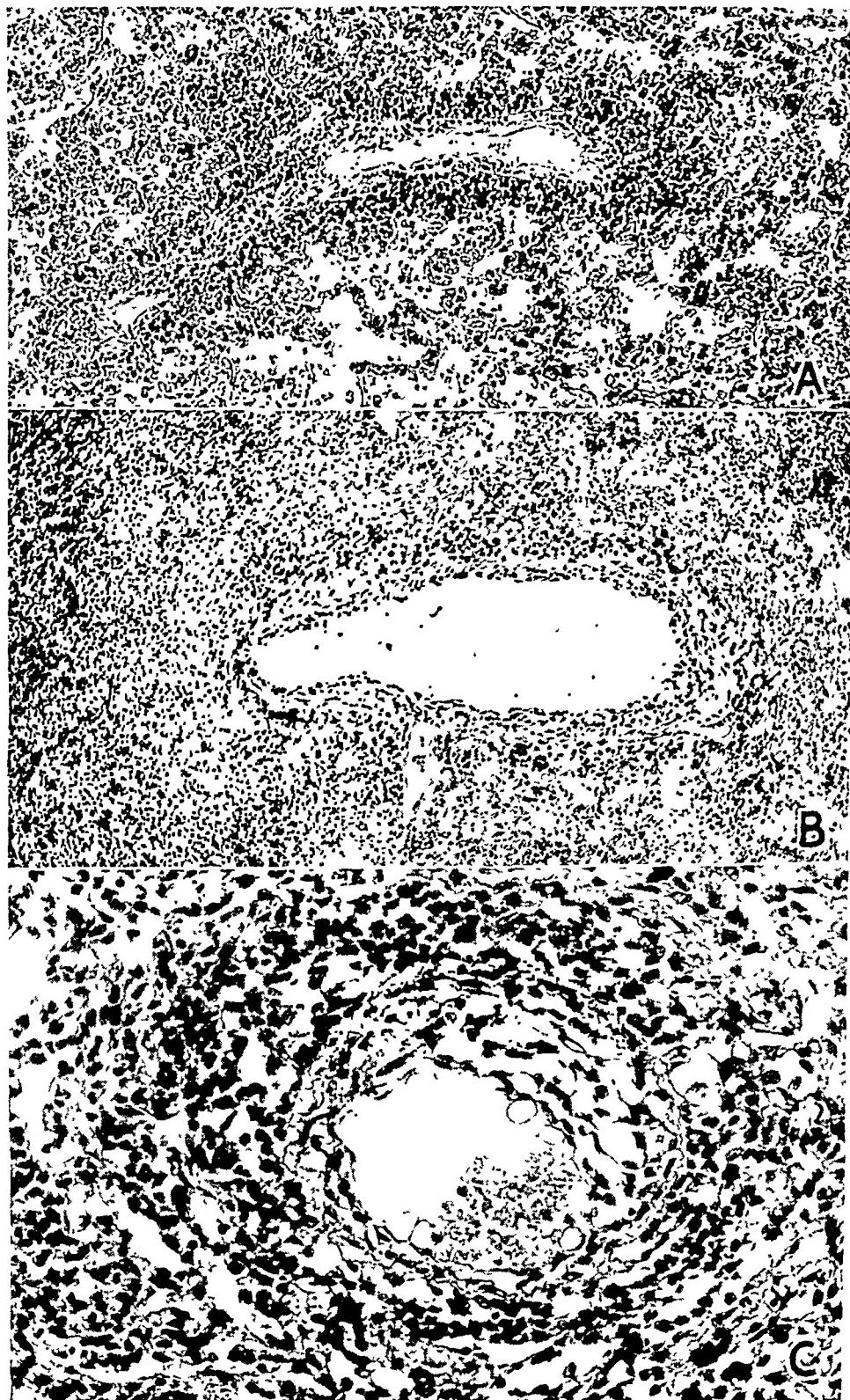


Fig. 5.—Different degrees of vascular changes. Section of the lung from an animal killed (A) two days, (B) three days and (C) four days following the "shocking" injection.

occupied an area from 1.5 to 3 cm. in diameter the exudate began to fade on the fourth day. In rabbits in which the lesion involved an entire lobe, pneumonic foci could be detected from eight to ten days after the injection of the "shocking" dose. In many animals the exudate disappeared entirely without leaving any traces, while in others, areas of organization were noticed on the fifth, sixth and seventh days after the "shocking" injection of the serum.

*Blood Vessels.*—The vascular system of the affected lung responded to the "shocking" dose as promptly as the parenchyma. The endothelial cells lining vessels of different caliber usually showed swelling and separation from the walls, singly and *en bloc*, thus adding to the overcrowded vascular bed containing excessive numbers of erythrocytes and granulocytes mingled with monocytes and occasional eosinophils. The walls of the vessels showed edema, and particularly remarkable was their early invasion by cells from within and without, leading to destruction of the architecture of the blood vessels. The almost complete occlusion of numerous small vessels by immigrated round cells and granulocytes has been described. Within a few hours after injection of the "shocking" dose of serum, numerous vessels showed fibrinoid changes of their walls. In many instances there was a proliferation of the intimal and subintimal layers causing thickening of the wall of the vessel, the newly formed tissue having a hyalinoid aspect. The adventitia of most of the vessels was surrounded by a cuff of mononuclear (monocytoid) cells, probably undifferentiated mesenchymal elements capable of transforming into monocytes and macrophages. Within the first few hours necrosis of the walls of the blood vessels (a vascularitis) was seen. The walls of the vessels were thoroughly infiltrated with myriads of cells, and it was surprising to find fibrin in the walls as well as in the lumens of the blood vessels. The presence of fibrin in the vascular lumens was a conspicuous trait in these cases. The picture of the pneumonic parenchyma and of the blood vessel shown in figure 4 is illustrative of the condition. There is an abundance of exudate made up of cells and serum. The perivascular lymphatics are distended and filled with cells. There is a great deal of fibrin. The wall of the vessel is permeated with cells which became "agglutinated," forming a nodule seen in the left upper segment of the picture (fig. 4). There is an abundance of fibrin in the lumen. Figure 5 shows changes in the wall of the vessel at different stages of the disease. In A there is a marked infiltration of the entire wall, somewhat resembling arteritis nodosa. In B (which shows a section of a lung removed from the thoracic cavity three days after the injection of the "shocking" dose), besides the cellular invasion of the wall of the vessel there is also a marked perivascular edema. In C the cellular infiltration forming a condition resembling a granuloma has finally led to complete dissociation of the wall of the blood vessel.

Infarctions extending to the pleura occurring early in the disease were conspicuous in many animals.

*Bronchi.*—The bronchial structures were found to be only mildly affected by the process, when compared with the gravity of the disease of the parenchyma and of the blood vessels. A peribronchial edema was present, but the bronchial walls showed no cellular infiltration, and the bronchial lining was intact.

#### COMMENT

The pulmonary reaction to small amounts of horse serum in rabbits sensitized with the same serum is remarkable in that there rapidly occurs an intense edema followed soon after by a spreading inflammation of the parenchyma and of the blood vessels, which in from twenty to

twenty-four hours involves one or more lobes of one lung and occasionally also the pleura. Similar or larger amounts of serum injected into the lungs of normal animals are usually lodged in the paravertebral regions of both lungs, which areas reveal a slight loss of their spongy appearance. In these animals the reaction of the lungs is slight; it occurs much later (about fifteen hours after the injection) and has a disseminated, insular character. In these cases the moderate exudate is composed of serum and granulocytes mingled with an occasional macrophage, and the lungs show no noticeable alteration of their configuration. In these animals the lungs regain their normal structure in from thirty to forty hours after the intratracheal injection, whereas in the sensitized animals signs of healing make their appearance in five or six days.

The mechanism of local allergic or anaphylactic inflammation studied by several investigators resulted in the following observation: When an antigen is introduced into man or lower animals it diffuses widely and rapidly into the circulation and the tissues, resulting in the formation of antibodies which can be detected *in vitro* by a specific precipitation test. But if animals are repeatedly treated with the same antigen (bacteria or foreign protein), the absorbing capacity of the antigen diminishes with each subsequent injection. Moreover, when the immunization is pursued, most of the antigen no longer disseminates, remaining at the site of injection, and, as in the experiments reported, causes a local acute exudative inflammation (Arthus' phenomenon). It is believed that local anaphylactic inflammation, like the general form of this manifestation, represents an altered capacity of the sensitized cells to react to the homologous antigen. The hypothesis was expressed that this type of inflammation occurs as the result of an interaction between the antigen and antibodies producing a toxic substance, a precipitin, according to Opie.<sup>4</sup>

The remarkable feature in the experiments here presented is that in a large percentage of cases the inflammation was confined to one lung, having a lobar distribution. The unilateral topography of the lesion is noteworthy because in control animals treated similarly with serum and dyes respectively the injected material invariably reached both lungs, setting up a mild inflammation in them.

Not only the topography but also the microscopic features of the inflamed lung are remarkable. Thus in a number of instances the pleura too was involved; the exudate composed in the first stages of serum and granulocytes was supplanted in from twenty-four to thirty hours by large mononuclear cells which crowded the alveoli and the septums. There occurred a widespread necrotizing arteritis, an abundant collection of fibrin and occasional infarctions. In animals killed about twenty-

4. Opie, E. L.: *J. Exper. Med.* **39**:659, 1924.

four hours after the intratracheal injection, the pulmonary lesion resembled in many respects that of acute lobar (genuine, fibrinous) pneumonia as seen in man.

In the period preceding Pasteur the cause of lobar or fibrinous pneumonia was attributed to chilling—frigus (“*Frigus unica pneumoniae causa [The sole cause of pneumonia is chilling]*”). With the advent of the bacteriologic era and the discovery of *Diplococcus lanceolatus*, “bacterium” took the place of “frigus,” and the notion was prevalent that the penetration of the micro-organism into the lower portion of the respiratory tract is alone responsible for the disease.

However, it soon became apparent that another factor besides the micro-organism is essential in setting up the malady. The view was expressed that the host, too, plays a rôle in the initiation of the pulmonary inflammation (“disposition” to the disease). This hypothesis was soon corroborated in experimental studies (Welch,<sup>5</sup> Beco,<sup>6</sup> and Rasquin<sup>7</sup>). In recent reports Stillman<sup>8</sup> stressed the observation that “Pneumococci which have reached the lung as a result of inhalation usually disappear within a few hours and give rise to no infection. . . . The experiments indicate that even in so susceptible an animal as the mouse, other factors than the presence of the pneumococci in the lung are necessary for infection.” Stuppy, Falk, and Jacobson<sup>9</sup> found that

the intratracheal inoculation of *Macacus rhesus* and *Cebus capucinus* monkeys with varying amounts of virulent type I pneumococci did not result in lobar pneumonia, although in most cases it caused an increase in temperature and a leucocyte reaction, with a pneumococcus septicemia. Recovery usually occurred within a week after inoculation.

Stuppy and Falk<sup>10</sup> noted that in rabbits

the intrabronchial insufflation with pneumococci of uniformly high virulence gave rise to a bronchopneumonia which usually caused death in two to five days, with septicemia and a generalized distribution of pneumococci in the lung. In some animals there occurred acute inflammation of the interstitial tissue of the lung and perivascular and peribronchial lymphangitis. Suppurative bronchitis and pleuritis were only occasionally seen.

I<sup>1</sup> have observed that a dose of pneumococci that kills a rabbit in approximately eighteen hours after injection into the blood stream causes the death of another rabbit in four days following intratracheal

5. Welch, William H.: Bull. Johns Hopkins Hosp. **3**:125, 1892.

6. Beco, L.: Arch. de méd. expér. et d'anat. path. **13**:51, 1901.

7. Rasquin, E.: Arch. de méd. expér. et d'anat. path. **22**:864, 1910.

8. Stillman, E. G.: J. Exper. Med. **38**:117, 1923.

9. Stuppy, G. W.; Falk, I. S., and Jacobson, M. A.: J. Prev. Med. **5**:81, 1931.

10. Stuppy, G. W., and Falk, I. S.: J. Prev. Med. **5**:89, 1931.

inoculation. On the other hand, a dose of this micro-organism causing the death of a hematogenously infected animal in from three to four days is harmless to an animal inoculated via the respiratory tract. In no instance could I induce an acute lobar inflammation of the lungs.

Briefly stated, lower animals resist the experimental induction of lobar pneumonia. The reason was not explained. Obscure, too, is the reason why in man the pneumococcus frequently causes a patchy (insular or miliary) bronchopneumonia scattered in both lungs, having a gradual onset, a protracted course and a "lytic" termination, while in other cases the same micro-organism initiates a disease with a rather sudden onset and abrupt termination ("cyclic" course), the lesion being lobar and unilateral.

Wadsworth<sup>11</sup> called attention to the fact that in animals that have been incompletely or partially immunized to the pneumococcus a subsequent infection with highly virulent pneumococci injected intratracheally causes a lesion akin to human lobar pneumonia. He noticed that in these cases the virulent micro-organism does not disseminate throughout the body and cause septicemia but remains fixed in the lungs, producing an acute lobar lesion.

Formerly observers were uncertain as to whether lobar pneumonia originates primarily in the respiratory organs or is a blood-borne condition which has "settled" in the lungs. The consensus today is that the disease is effected through inhalation, by way of the upper respiratory tract, of *Diplococcus lanceolatus*, rarely of *Diplococcus mucosus*, *staphylococci* or *streptococci*. The disease starts most likely in the parenchyma of the lung, namely, at the periphery of the acinus (and not in the bronchioles), from which it spreads toward the surrounding parenchyma *per continuitatem* by way of the lymphatics, toward the hilar lymph nodes, and also possibly by way of the bronchioles (canalicular spread), stimulated by coughing. The process, as it is known, usually involves one lung, and, as a rule, is confined to one lobe. From this observation the inference was that the rapidly spreading inflammation limited to one lung results probably from the fixation *in situ* of inhaled pneumococci in the previously sensitized pulmonary tissue, or that it is a local allergic manifestation. It was pointed out that lobar pneumonia occurs rarely, if at all, in babies under 5 months of age. that cases of fibrinous (genuine) pneumonia that have been reported to have occurred in infants in the first few months of life were usually traced to a transplacental transmission of the hypersensitivity and of the disease from the mother at the time of delivery. It has been stated that the resistance of babies to genuine pneumonia is due to the fact that they had no opportunity of becoming infected and are therefore

11. Wadsworth, A.: Am. J. M. Sc. 127:851, 1904.

relatively immune. Thus the "virgin" lung of the baby, like that of the normergic lower animal, rapidly disposes of the pneumococcus when it is inhaled for the first time without undergoing an acute lobar inflammation. On the contrary, in a lung that has been sensitized on previous occasions a minimal number of pneumococci is likely to cause a massive involvement.

I have quoted the experiments by Wadsworth to this effect. His work was repeated with contradictory results. For example, Stillman and Branch<sup>12</sup> found that "mice which have been partially immunized by previous inhalations of living or killed pneumococci and while alcoholized are exposed to an atmosphere of virulent pneumococci often develop a pneumococcus lobar pneumonia." Stuppy, Cannon and Falk<sup>13</sup> succeeded in inducing an acute exudative lesion in the lung of partially immunized rabbits.

Sharp and Blake<sup>14</sup> noted that in rabbits rendered hypersensitive to the pneumococcus, a subsequent intrabronchial insufflation of this micro-organism caused an acute inflammatory reaction of the lung in a number of instances. Lindau<sup>15</sup> found that "sterile pneumococcus autolysate, injected intratracheally into guinea-pigs with positive skin test, caused pulmonary reactions with lobar tendency." Lauche<sup>16</sup> detailed in many articles the allergic conception of genuine pneumonia of which he is a follower. In his studies of a large number of pneumonic human lungs Loeschke<sup>17</sup> found that the inhaled pneumococci "set up a hyperergic reaction in a sensitized allergic organism." According to the observations of this author the hyperergic (allergic) reaction causes at the point of inoculation an abundant edema in which the pneumococci proliferate with great rapidity. The exudate rich in pneumococci spreads throughout the lung by way of the interalveolar pores, setting up in the newly invaded alveoli an allergic type of response.

It is noteworthy that by intratracheal injection of egg albumin or of pneumococcus protein Julianelle and Roads<sup>18</sup> succeeded in inducing an inflammatory reaction in the lungs of rabbits previously inoculated with the respective antigen. They also found that a similar reaction occurs following an intratracheal injection of pneumococcus protein into the lungs of rabbits previously inoculated with heat-killed suspensions of the bacteria. However, their experiments "failed to show that hyper-

12. Stillman, E. G., and Branch, A.: *J. Exper. Med.* **54**:623, 1931.

13. Stuppy, G. W.; Cannon, P. R., and Falk, I. S.: *J. Prev. Med.* **5**:97, 1931.

14. Sharp, E. A., and Blake, F. G.: *J. Exper. Med.* **52**:501, 1930.

15. Lindau, A.: *Acta path. et microbiol. Scandinav.* **10**:1, 1933.

16. Lauche, A., in Henke, F., and Lubarsch, O.: *Handbuch der speziellen pathologischen Anatomie und Histologie*, Berlin, Julius Springer, 1928, vol. 3, p. 915.

17. Loeschke, H.: *Beitr. z. path. Anat. u. z. allg. Path.* **86**:201, 1931.

18. Julianelle, L. A., and Roads, C. P.: *J. Exper. Med.* **55**:797, 1932.

sensitiveness to pneumococcus or its products influences the occurrence or character of the lesion in the lungs in artificial pneumococcus infection."

In a recent article Cole<sup>19</sup> treated another aspect of the problem. He wrote:

With increasing experience, it becomes more and more evident that pneumonia rarely arises primarily through infection with the organisms responsible for the pneumonic lesions, but that for these organisms to induce infection the soil must be prepared, probably by some other less serious infection, usually one involving the upper respiratory tract.

This "dualistic" conception of the pathogenesis of lobar pneumonia was supported by "careful investigation which shows . . . that many patients with pneumonia exhibit symptoms referable to the upper respiratory tract for a longer or shorter period before the onset of the disease itself" (Cole). Indeed, it is possible that the preceding "less serious infection" produces a nonspecific sensitization of the lung toward the pneumococcus. It may also be that the "upper respiratory" disturbances preceding lobar pneumonia are none other than premonitory symptoms of the disease "smoldering" in the lung, for the clinical onset of the disease in genuine pneumonia, as in other morbid conditions, does not coincide with the moment of infection. The abrupt onset of lobar pneumonia seen in many patients usually coincides with a rather considerable involvement of the lung and often also of the pleura. It should be remembered that the lung remains "solid" for a considerable length of time after the apparent (clinical) cure of the disease.<sup>20</sup>

In this respect it is of interest to note that the reaction produced by the final injection of the protein into sensitized animals was designated by writers as "shocking," "fulminating" and "explosive," the inference being that it is akin to the general anaphylactic shock, which was compared by Besredka to an explosion produced by an abrupt pouring of water into sulphuric acid. Indeed, if tissues are examined from twenty-four to forty-eight hours after the final injection of the antigen, the impression is gained that an "explosion" has occurred because of the profound alteration of the tissues in and around the area of injection. In reality it takes from fifteen to twenty-four hours for the local allergic inflammation to attain its height. This holds true of the lungs as well as of the cutaneous lesion produced by Arthus.

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19. Cole, Rufus: Canad. M. A. J. **30**:237, 1934.

20. Since "time immemorial" alcohol was considered an important factor in the contraction of lobar pneumonia. This was not investigated seriously; it has never been established that the disease occurs with greater frequency in alcoholics. Moreover, as in the case of the "upper respiratory" disturbances, it is not unlikely that the "intoxication" merely coincided with the onset of the malady at the time when it showed no demonstrative clinical manifestations.

It is important to stress that, unlike experimental infection, natural infection by way of the respiratory tract usually occurs with small amounts of bacteria which cause no disease in the normergic lower animal and in all likelihood in the normergic person. But in the allergic subject small quantities of bacteria produce the hyperergic type of inflammation, just as in sensitized animals small amounts of serum produce the acute lesion heretofore described. Moreover, as stated, a general systemic dissemination is prevented; that is, the organism remains fixed *in situ*.

#### SUMMARY

When an antigen is repeatedly injected subcutaneously into an animal, its absorbing capacity diminishes with each subsequent injection. Moreover, in these cases the last injection (usually the fifth or sixth) produces an acute exudative inflammation locally, looked on as an anaphylactic phenomenon; it is assumed that it occurs as a result of a combination of antigen and antibodies.

In the experiments reported in this article rabbits were sensitized by repeated intraperitoneal injections of horse serum, the last injection (defined as "shocking") being introduced into the lungs via the trachea.

A study of the lungs of these animals showed that: 1. The intratracheal ("shocking") injection of the heterologous serum soon produced an acute exudative, rapidly spreading inflammation in the lungs. 2. In a high percentage of the animals studied the lesion at its height was confined to one lung, having a diffuse lobar distribution. 3. The gross and microscopic aspects of the pulmonary lesion resembled in many ways acute lobar (fibrinous or genuine) pneumonia as seen in man.

The nature of the latter disease was analyzed, and the hypothesis was expressed (in accord with the views of earlier observations) that in this malady the character of the lesion in the lung probably depends on a state of local pulmonary hypersensitivity (allergy, hyperergy).

The similarities between the lesions in the lungs in genuine pneumonia in man and those observed in the experiments with horse serum reported in this article (which should be regarded as "model" infections) favor this hypothesis.

# EFFECT OF PARATHYROID EXTRACT ON THE BONES OF THE HYPOPHYSECTOMIZED RAT

A HISTOLOGIC STUDY

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Jaffe and his co-workers<sup>1</sup> showed that large doses of parathyroid extract lead to osteoclastic bone resorption in various experimental animals. Later Johnson<sup>2</sup> pointed out that he sometimes observed the formation of osteoid tissue in the bones of animals given this preparation. We have never seen osteoid formation as a result of over-dosage of parathyroid extract in this laboratory, but we have regularly been able to stimulate the formation of fully calcified normal bone tissue by the chronic administration of this extract to rats. Some difference in the diet or in the caging conditions may perhaps account for this discrepancy between Johnson's observations and ours. Our studies of the more minute morphogenetic action of parathyroid extract on the osseous tissue have shown that it is necessary to differentiate between two entirely different types of bone reaction to the extract: the osteoclastic and the osteoblastic reaction.<sup>3</sup> Large doses of parathyroid extract lead to the proliferation of osteoclasts and simultaneously to bone resorption. When the daily administration of the extract is continued, the experimental animal, if it survives this stage, becomes gradually more and more resistant, and its clinical condition improves in spite of the continued treatment. During this stage of increased resistance the bone shows a disappearance of osteoclasts and proliferation of numerous osteoblasts, with formation of entirely normal, calcified new bone. If the treatment is started with a very small dose, one may even stimulate the formation of new bone tissue immediately, without a preliminary stage of bone resorption.<sup>3a</sup>

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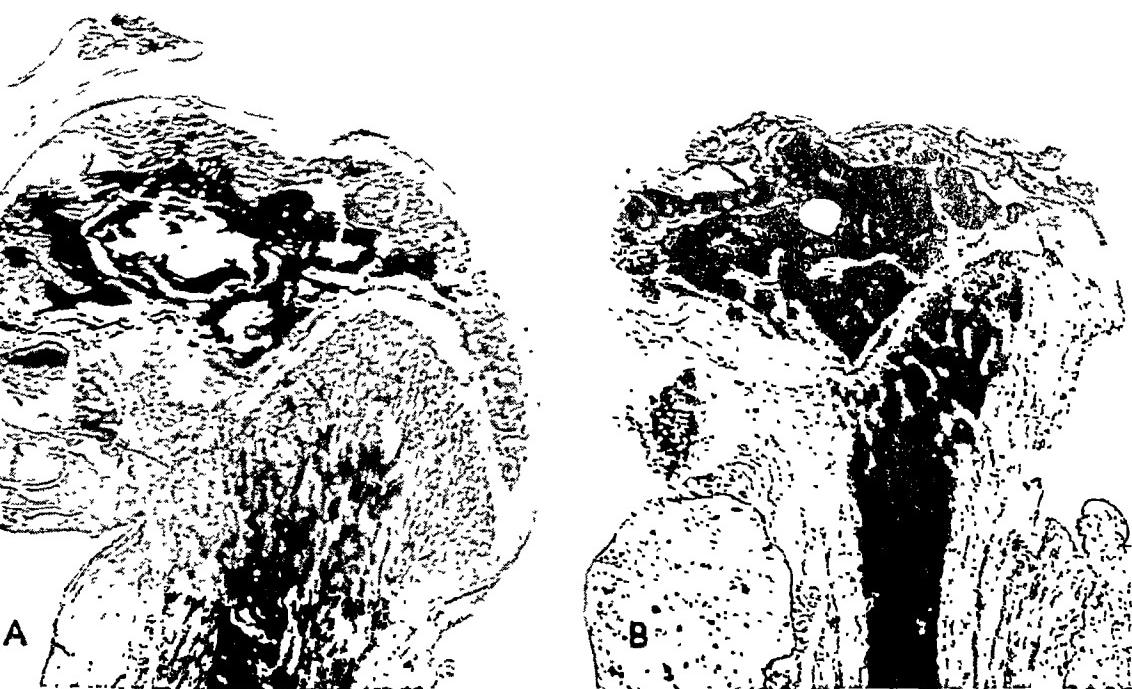
From the Department of Biochemistry, McGill University.

1. Jaffe, H. L.; Bodansky, A., and Blair, J. E.: Klin. Wchnschr. **9**:1717, 1930; J. Exper. Med. **55**:695, 1932.

2. Johnson, J. L.: Am. J. M. Sc. **183**:769, 1932.

3. Selye, H.: (a) J. A. M. A. **99**:108, 1932; (b) Endocrinology **16**:547, 1932.

Since the pituitary growth hormone is considered to be essential for the growth of the skeleton and hypophysectomy stops growth, we have been interested to see whether the formation of osseous tissue by treatment with parathyroid extract, as observed in our previous experiments, is also dependent on the presence of the hypophysis. In order to study this question, we hypophysectomized twenty male rats, from 40 to 50 days of age, thirteen of which we treated with graded daily doses of parathyroid extract, starting with 1 unit on the first day and increasing the dose by 1 unit every day until the eighth day, after which they received 8 units daily. The remaining seven animals were used as controls, and intact treated and untreated animals of similar



*A* shows the lower end of the femur of an albino rat twenty-six days after hypophysectomy. Note the poor development of the trabeculae in the subepiphyseal region. *B* shows the lower end of the femur of an albino rat hypophysectomized twenty-six days before and since then treated with small doses of parathyroid extract.

age were also studied. Two animals of each group were killed on the eighth day of treatment; at this period the hypophysectomized rats showed marked bone resorption with the formation of numerous osteoclasts, while the normal animals receiving the same treatment already showed new bone formation and proliferation of osteoblasts. The remaining animals of both groups were killed in pairs between the twelfth and the twenty-fifth day of treatment. While the bone apposition was always much more marked in a normal animal than in its hypophysectomized control, it became obvious that in a later stage—

from the fourteenth day on—the hypophysectomized animals also began to show proliferation of osteoblasts and formation of new bone. This became marked in animals killed during the third week and later.

From these experiments we conclude that parathyroid extract may stimulate the formation of bone tissue even in the absence of the hypophyseal growth hormone.

In this connection we should like to emphasize a few facts which we believe are important for the understanding of the mechanism of the action of the growth hormone. We have observed that compensatory hypertrophy of the remaining kidney occurs in hypophysectomized rats after unilateral nephrectomy; furthermore, Dr. Jeffers has found in this laboratory that mitotic figures are plentiful in the mammary glands of hypophysectomized rats at the time of parturition. It is also well known that the proliferation of fibroblasts and epithelial cells in wound healing is not seriously interfered with by hypophysectomy, and that transplanted tumors grow, though less rapidly than usual, in hypophysectomized rats. All of these observations show that the proliferation of cells and the growth of individual organs are not necessarily dependent on the growth hormone. That animals may survive hypophysectomy for several years shows distinctly that cells of the various organs which died during this period have been replaced by new ones. The present paper shows that even a generalized growth (although not increase in length) of the osseous system may be produced in the absence of the pituitary growth hormone.

We conclude that the growth of the various organs in themselves is largely independent of the hypophysis. Apparently the function of the pituitary growth hormone is only to permit enlargement of the size of the body as a whole, with a harmonious and proportional increase in the size of all the organs.

Eli Lilly & Co. supplied the parathyroid extract used in these experiments.

## DI-NITROPHENOL

STUDIES OF BLOOD, URINE AND TISSUES OF DOGS ON CONTINUED  
MEDICATION AND AFTER ACUTE FATAL POISONING

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D. A. WOOD, M.D.

AND

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SAN FRANCISCO

In connection with the therapeutic use of 2, 4, di-nitrophenol<sup>1</sup> it appeared desirable to determine what changes, if any, in vital functions and organs might be produced by prolonged administration of doses comparable to the clinical ones and by acutely fatal doses. Tainter and Cutting<sup>2</sup> have reported briefly the somewhat indefinite histologic changes observed after the injection of large or fatal doses into dogs for periods of about one month. Schulte and Tainter<sup>3</sup> observed no harmful effects on the function of the kidneys in rabbits medicated for seventy-seven days. In the present report, the effects of nonfatal doses administered to dogs daily for six months are described.

### METHODS

Nine adult male dogs were kept in large cages and fed all they would eat of a balanced commercial dog food. In addition, they were given cod liver oil and frequent exercise outdoors in the sun. The experiment was started after a period of quarantine of one month. Each week, three of the dogs were weighed and placed in separate metabolism cages, and quantitative collections of urine were made for twenty-four or more hours. During this period, samples of blood of from 10 to 15 cc. were obtained from each dog, using a vein of the leg. The urine was examined for sugar by Benedict's qualitative method and for albumin by the

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The studies were supported in part by a grant from the Rockefeller Fluid Research Fund of the Stanford University School of Medicine.

From the Departments of Pharmacology, Medicine and Pathology, Stanford University School of Medicine, San Francisco, and the Santa Clara County Hospital, San Jose, Calif.

1. Cutting, W. C.; Mehrtens, H. G., and Tainter, M. L.: J. A. M. A. **101**: 193, 1933. Tainter, M. L.; Stockton, A. B., and Cutting, W. C.: ibid. **101**:1472, 1933. Cutting, W. C., and Tainter, M. L.: ibid. **101**:2099, 1933.

2. Tainter, M. L., and Cutting, W. C.: J. Pharmacol. & Exper. Therap. **49**: 187, 1933.

3. Schulte, T. L., and Tainter, M. L.: Proc. Soc. Exper. Biol. & Med., **31**: 1163, 1934.

quantitative method of Shevky and Stafford.<sup>4</sup> The samples of blood were used for determination of: the hemoglobin content in grams per hundred cubic centimeters, using the acid hematin method and a fixed glass standard in the Klett colorimeter; the red, white and differential cell counts by the usual clinical methods; the blood urea by the urease method, and the icteric index of the plasma against a fixed glass standard in the colorimeter. Before reading the icteric index, the di-nitrophenol in the serum was decolorized by dilution with tenth-normal hydrochloric acid. Preliminary observations showed that the acid did not alter the color of the bile pigments in the serum and hence did not change the icteric index when no drug was present.

After a control period of six weeks, dogs A, D and G were designated as control animals; B, E and H were given 5 mg., and C, F and I, 10 mg., of di-nitrophenol per kilogram of body weight, orally in capsules, daily for six days in each week. The average daily therapeutic dose for man is about 3 mg. per kilogram; so the dogs received up to about three times the usual clinical dose. Dog E, receiving 5 mg., was killed in a fight in the middle of the experiment. The others remained in apparent good health during the twenty-seven weeks that the drug was given. At the end of the twenty-fourth and of the twenty-seventh week, the oxygen capacity of the blood was determined by means of the device described by Anrep and Harris,<sup>5</sup> and the fragility of the red blood cells was determined by their resistance to hypotonic salt solutions.<sup>6</sup> Then the experiment was discontinued, and the dogs were killed for histologic study of the tissues. Each dog was killed by injecting a solution of a cyanide salt intravenously, and a complete necropsy was performed. The brain was removed at the moment of death, and one half was fixed in alcohol and the other half in a neutralized solution of formaldehyde made isotonic with potassium bromide.

#### CLINICAL RESULTS

The results may be discussed according to the tissue, fluid or function studied.

*Body Weight.*—The average initial weight of the three control dogs was 8.3 Kg., and the average weight at the end of the experiment was 9 Kg., a gain of 0.7 Kg. The two dogs receiving 5 mg. daily, which survived for six months, weighed at the beginning of administration of the drug an average of 9 Kg., and at the end, 8.4 Kg., a loss of 0.6 Kg. The three dogs given doses of 10 mg. started with an average weight of 12.5 Kg. and ended with an average weight of 12.8 Kg., a gain of 0.3 Kg. These results show that there were no important changes in body weight as a result of the continuous administration of di-nitrophenol during six months, under the conditions employed.

*Urinary Protein and Sugar.*—No sugar was found in the urine of any of the dogs throughout the entire period of medication. The changes in the output of urinary protein in twenty-four hours are shown graphically in chart 1. The highest daily output during the control period was 75 mg. Dog H, given doses of 5 mg. of di-nitrophenol, excreted up to a peak of 142 mg. of albumin during the twelfth week of medication, but the excretion fell to the control level of 12 mg.

4. Shevky, M. C., and Stafford, D. D.: Arch. Int. Med. 32:222, 1923.

5. Anrep, G. V., and Harris, D. T.: Practical Physiology, London, J. & A. Churchill, 1923, p. 99.

6. Todd, J. C., and Sanford, A. H.: Clinical Diagnosis by Laboratory Methods, ed. 6, Philadelphia, W. B. Saunders Company, 1928, p. 363.

at the end of the experiment, when the drug was still being given. Dogs F and I, given doses of 10 mg. of di-nitrophenol, showed a similar increase in the protein output at the same time, but a return to normal levels occurred while the drug was still being given. At the time of the greatest excretion of albumin the urine gave only a faintly positive reaction with heat and acetic acid; so the changes, although

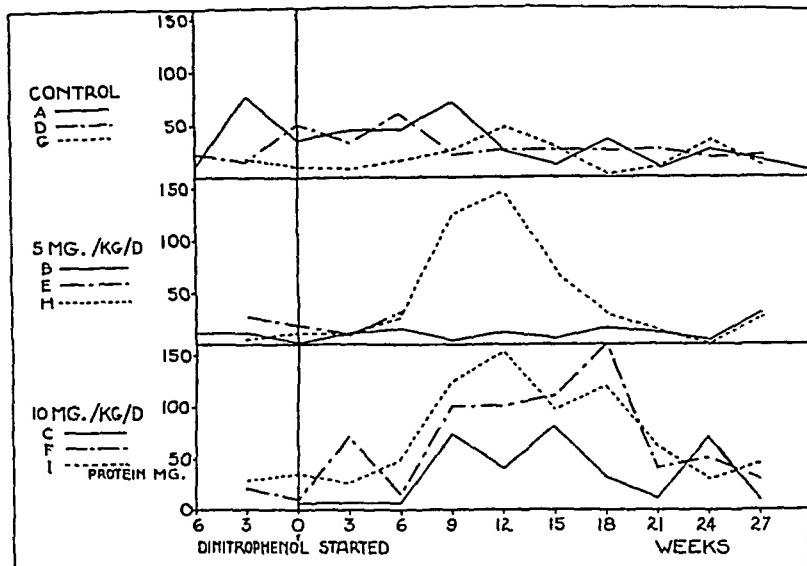


Chart 1.—Effects of di-nitrophenol administered orally on amount of albumin in the urine of dogs in twenty-four hours. In this and the following charts, each line represents one dog, and Mg./Kg/D indicates the dose of di-nitrophenol in milligrams per kilogram per day.

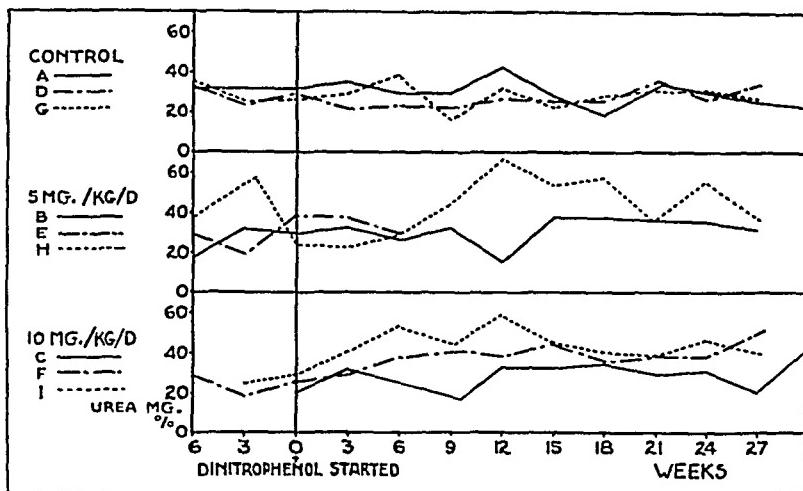


Chart 2.—Effects of di-nitrophenol administered orally on the blood urea concentration of dogs.

definite, were not of sufficient magnitude to be important in the absence of other evidence of renal damage. The fact that the final values were all in the normal range also indicates that the di-nitrophenol did not cause progressive damage of the kidneys but that the temporary increase in the albumin content was possibly due to some intercurrent factor.

*Blood Urea.*—The absence of significant renal damage was confirmed by the practically negative results of determinations of the blood urea. In chart 2 it may be seen that there was no consistent change in the level of this blood constituent during the period of medication. Hence, the kidneys were not functionally damaged, according to this test.

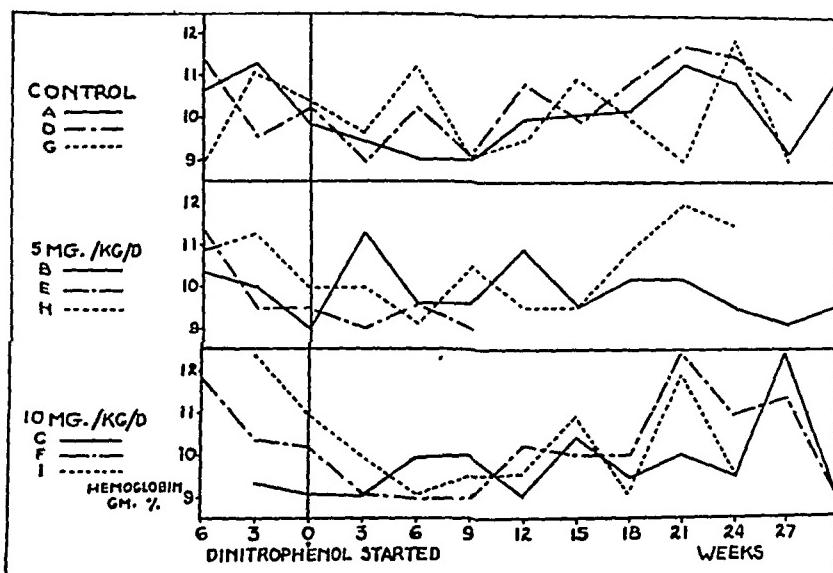


Chart 3.—Effects of di-nitrophenol administered orally on the hemoglobin content of the blood of dogs.

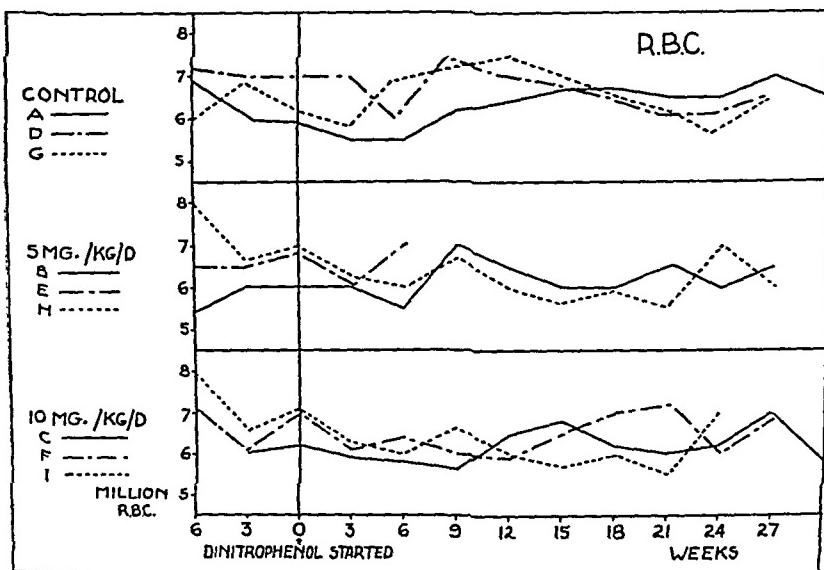


Chart 4.—Effects of di-nitrophenol administered orally on the red blood cell count of dogs.

*Hemoglobin and Red Blood Cells.*—The hemoglobin content of the blood was found to be rather variable. This was undoubtedly due partly to difficulties in the method, since some trouble was experienced at times because of a poor color match with the standard. The average results (chart 3) indicated no significant change in the concentration of hemoglobin. Counts of the red blood cells also showed no

significant change (chart 4). At the end of the twenty-fourth week of the administration of di-nitrophenol and again at the end of the twenty-seventh week, the oxygen capacity and the fragility of the red cells were determined. The control dogs gave an average value of 14.5 per cent by volume for oxygen capacity;

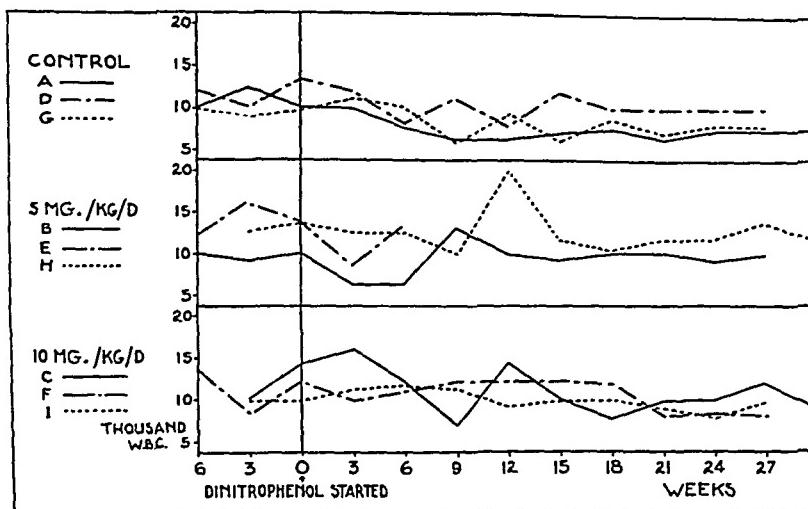


Chart 5.—Effects of di-nitrophenol administered orally on the white blood cell count of dogs.

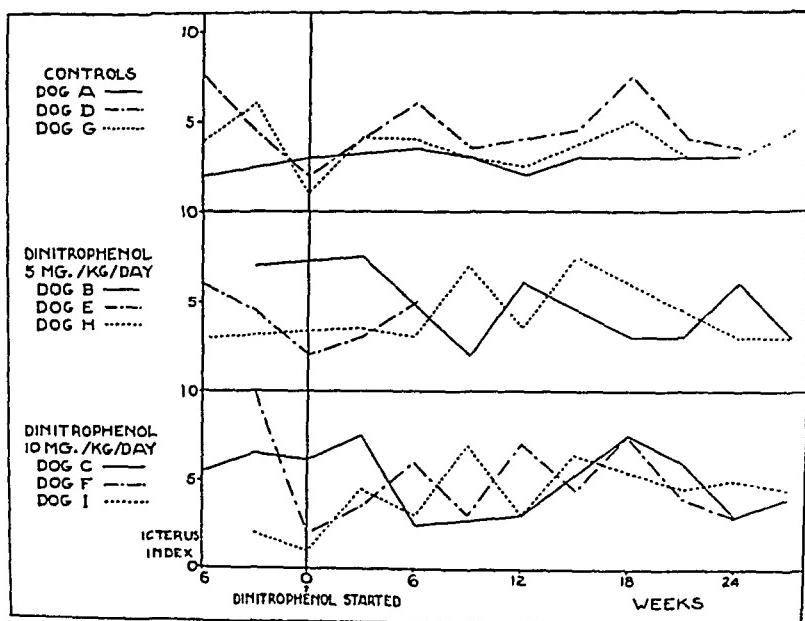


Chart 6.—Effects of di-nitrophenol administered orally on the icterus index of the blood serum of dogs.

the dogs given 5 mg. doses, 15.3 per cent, and those given 10 mg. doses, 16.1 per cent. The individual values ranged between 13.5 and 18 per cent. Accordingly, it appears that di-nitrophenol, if anything, increased slightly the average oxygen capacity of the blood; and, what is more important, there was no decrease.

In the fragility tests made at the same time, the results were very uniform. In all the dogs, hemolysis began with concentrations of the salt of 0.462 and 0.423 per cent and was complete with concentrations of 0.336 and 0.295 per cent. There was thus no alteration in the fragility of the red cells, as shown by their resistance to laking by hypotonic solutions.

*White Blood Cells.*—The total number of white blood cells remained unchanged throughout (chart 5). Differential counts from smears failed to show any alteration in the proportions of eosinophils, basophils, lymphocytes or large mononuclear or polymorphonuclear neutrophils. There were no abnormal cells present, as judged from shape, size or staining characteristics. The blood picture, therefore, remained normal throughout the six months of medication.

*Icteric Index.*—The theoretical possibility of damage to the liver led to observations of the icteric index. The average initial value for the control dogs was 3.7, and at the end of the experiment the average value was 3.3. The dogs receiving di-nitrophenol gave an average of 3.8 at the beginning of the administration and of 3.5 at the end. In no instance did the index go above the normal range during the administration of di-nitrophenol (chart 6). Therefore, this test revealed no evidence of damage to the liver from the drug.

*Comment.*—The tests described failed to show that six months of continuous oral administration of di-nitrophenol to dogs, in doses three or more times the clinical, produced any pathologic changes, unless such changes were indicated by the small increases in the amount of albumin in the urine. However, it is believed that the albuminuria was coincidental, since there was a return to normal values during medication. Further evidences of pathologic changes, therefore, were sought by histologic study of the tissues.

The dogs were killed by injecting a solution of a cyanide salt intravenously, and necropsies were performed immediately. Routine gross and microscopic examinations were made of the brains, tissues and organs from each dog. No essential changes could be detected in any of the animals which were not present in one or more of the controls.

#### CHANGES OBSERVED POST MORTEM IN THE BRAIN

In addition to the brains of the dogs already discussed, those of three other dogs which were acutely poisoned by injecting fatal doses of di-nitrophenol subcutaneously were studied. Two of these dogs had received 30 mg. of di-nitrophenol per kilogram of body weight, and one had been given 20 mg. per kilogram; all died within three or four hours from hyperpyrexia.

Immediately after death, the brains of both the chronically and the acutely poisoned animals were removed, and one half of each was fixed in 96 per cent alcohol and the other half in a 4 per cent solution of formaldehyde. After fixation, blocks were removed from the frontal, motor, parietal, occipital and temporal regions, including the subcortical centers, as well as from the cerebellum, the medulla oblongata and the cervical portion of the cord.

The alcohol-fixed material was stained en bloc with methylene azure, embedded in paraffin and cut in sections from 15 to 20 microns thick. Paraffin sections of alcohol-fixed material were also stained with coelestin blue-eosin, chromium alizarine cyanin and alum-gallamin blue.<sup>7</sup> The fibrous and protoplasmic glia were stained by Cajal's method, and the microglia by Hortega's method combined with Herxheimer's fat stain. Spielmeyer's and Bielschowsky's methods were used for staining the myelin sheaths and axis-cylinders.

Sections of the various regions of the cortex of the acute and chronically poisoned animals, stained with methylene azure en bloc, showed the nerve cells intact, with the exception of a few sclerotic cells scattered throughout the cortical layers. Sclerotic cells were also found in the control animals and were probably due to senile changes. Hemorrhages and cellular infiltrations were found in dog H only, and were undoubtedly of infectious origin. The glia cells were unchanged, no regressive or progressive changes being demonstrable. A slight perivascular fatty infiltration was found in all animals, including the controls. The nerve and glia cells of the striatum and pallidum, the ganglion cells of the cornu ammonis and the Purkinje cells of the cerebellum and nucleus dentatus were all intact, as were the nuclei of the medulla oblongata. The cervical portion of the cord was normal. No changes could be demonstrated in the myelin sheaths or in the axis-cylinders.

#### CHANGES OBSERVED POST MORTEM IN OTHER TISSUES

The other tissues from the dogs used in the chronic feeding experiment were subjected to careful gross and microscopic examination. The specimens were fixed in Orth's and Zenker's fluids. Frozen sections were made of each tissue and stained with hematoxylin and sudan III for the demonstration of fatty changes. Parts of each tissue were embedded in paraffin, cut in sections 5 microns in thickness and stained with hematoxylin and eosin and hematoxylin and Van Gieson's stain. Bone marrow sections were stained with Giemsa's stain and hematoxylin and eosin.

The following protocols are arranged according to the dosage of di-nitrophenol.

*Dogs Given 10 Mg. of Di-nitrophenol per Kilogram.*—Dog C.—Grossly all the organs were essentially normal.

The results of microscopic examination were as follows: The liver was moderately congested throughout. The central veins and sinusoids were slightly dilated. The neighboring liver cells were slightly atrophic but showed normal cytoplasmic and nuclear staining. No fatty changes were observed. One testicle had a normal structure. In the other there was atrophy of the tubules with absence of spermatids and spermatocytes. There were no demonstrable changes in the interstitial cells. The lungs showed moderate congestion of the alveolar and septal capillaries and blood vessels. The heart presented slight atrophy of the muscle fibers. The thyroid gland was moderately congested. The acini were of normal size and shape and were filled with colloid which stained uniformly with eosin. The lymph nodes showed slight lymphoid hyperplasia. The islands of Langerhans were numerous but showed no abnormalities. The reticulum of the pulp of the spleen showed slight fibrous thickening and contained small amounts of finely granular brown pigment. The cells of the suprarenal cortex were heavily laden with lipoid. The kidneys were normal, with no changes in the tubules. The epididymis, stomach, intestines, bone marrow and skeletal muscles were normal.

Dog F.—Grossly all the organs were essentially normal.

7. Proescher, F., and Arkush, A. S.: Stain Technol. 3:28, 1928, Einarson, L.: Am. J. Path. 8:293, 1932.

The results of microscopic examination were as follows: The liver showed normal architecture throughout. The central venules and sinusoids were moderately congested, but no fatty or degenerative changes were observed. In the lungs there was marked capillary congestion. The lymph nodes showed slight lymphoid hyperplasia. The islands of Langerhans were rather numerous and moderately congested. Scattered throughout the pulp of the spleen were small amounts of brown, granular pigment. The malpighian bodies were rather prominent, with their centers less dense than their peripheries. The cells of the suprarenal cortex were heavily laden with lipoid. There was moderate congestion throughout the suprarenals. The kidneys showed occasional tiny cortical scars and occasional small groups of round cells. In some of the convoluted tubules there was slight to moderate cloudy swelling of the lining epithelium. A few hyaline casts were contained in some of the collecting tubules. The glomeruli were normal. The kidneys were moderately congested throughout. The testicles, epididymis, heart, thyroid gland, stomach, intestines, bone marrow, skeletal muscles and parathyroid glands were normal.

**DOG I.**—Grossly all the organs were essentially normal.

The results of microscopic examination were as follows: The liver was markedly congested throughout. All the central sinusoids were greatly distended with red blood corpuscles. In some areas the central lobular cells showed marked atrophy. No fatty or hydropic degenerative changes were present. In the lungs the alveolar capillaries were markedly congested. In the thyroid gland nearly all the acini were filled with colloid, and many were rather large. The parathyroid glands were moderately congested. The lymph nodes showed slight lymphoid hyperplasia. Scattered throughout the pulp of the spleen were small amounts of brown, granular pigment. The malpighian bodies were prominent. The trabeculae showed slight fibrous thickening. The cells of the suprarenal cortex were heavily laden with lipoid. The kidneys showed marked congestion of the glomerular capillaries. In some of the convoluted tubules there was slight cloudy swelling of the epithelium. A few hyaline casts were contained in some of the collecting tubules. The testicles, epididymis, heart, stomach, intestines, bone marrow and skeletal muscles were normal.

*Dogs Given 5 Mg. of Di-nitrophenol per Kilogram.*—**DOG B.**—Grossly all the organs were essentially normal.

The results of microscopic examination were as follows: The liver was moderately congested. The lungs showed moderate congestion of the alveolar capillaries. The thyroid gland was normal except that in a few acini the colloid stained bluish with hematoxylin instead of pinkish with eosin. The germinal follicles of the lymph nodes were well developed. One lymph node showed chronic inflammation of mild degree. The islands of Langerhans were rather numerous and normal except for moderate congestion. The spleen was normal, and no pigment was present. The malpighian bodies were of normal size and appearance. In the cortex of the kidneys occasional tiny scars were present and were associated with small collections of round cells. Some of the convoluted tubules showed slight cloudy swelling of the lining epithelium. No casts were observed. The submaxillary salivary glands, testicles, epididymis, heart, suprarenal glands, stomach, intestines, bone marrow, skeletal muscles and prostate were normal.

**DOG H.**—Grossly all the organs were essentially normal.

The results of microscopic examination were as follows: The liver was moderately congested, and slight fatty infiltration was observed. The lungs were moderately congested throughout. The spleen showed a small amount of brown

granular pigment scattered throughout the pulp. The malpighian bodies were rather prominent and showed slight lymphoid hyperplasia. The cells of the suprarenal cortex were heavily laden with lipoid. The medullary portion of the suprarenals was moderately congested. The kidneys showed spontaneous nephritis of mild degree. A few of the glomeruli showed intraglomerular fibrosis of the tufts with adhesions to Bowman's capsule. These glomeruli were infiltrated by a few round cells. In scattered groups of convoluted tubules there was fatty degeneration of the lining epithelium. In these areas the cells were desquamating and also showed considerable cloudy swelling. The interstitial tissue showed slight fibrous thickening. A few of the tubules contained cellular and hyaline casts. As a whole, however, the renal changes were limited in extent. The bone marrow showed moderate hyperplasia. The endothelial macrophages lining the sinuses were large and contained phagocytosed cellular débris. The testicles, epididymis, heart, thyroid gland, parathyroid glands, lymph nodes and skeletal muscles were normal. This was the animal which showed alterations in the excretion of albumin and cellular infiltrations in the brain. It is likely that all the changes noted were due to a previous attack of distemper and were in no way associated with the administration of di-nitrophenol.

*Control Dogs Killed at the End of Six Months (No Di-nitrophenol).*—Dog A.—Grossly all the organs were essentially normal.

The results of microscopic examination were as follows: In the liver a few small periportal collections of round cells were present. A few of the hepatic cells possessed foamy cytoplasm and with fat stains showed small areas of fatty degeneration. The lungs showed moderate congestion of the alveolar capillaries. In the spleen there was considerable fibrous thickening of the reticulum as well as of the trabeculae. Moderate amounts of brown granular pigment were deposited in the pulp. The malpighian bodies were normal. The cells of the suprarenal cortex were moderately laden with lipoid. The kidneys showed occasional tiny cortical scars and small groups of round cells. Some of the glomeruli showed slight periglomerular fibrosis. The testicles, epididymis, heart, thyroid gland, parathyroid glands, lymph nodes, bone marrow, stomach, intestines, skeletal muscles, submaxillary salivary glands and prostate were normal.

Dog D.—Grossly all the organs were essentially normal.

The results of microscopic examination were as follows: The liver was moderately congested. Many of the central lobular sinusoids were widely dilated and congested, and there was atrophy of the neighboring liver cells. The lungs were moderately congested throughout. In the pulp of the spleen small amounts of brown granular pigment were present. The cells of the suprarenal cortex were moderately laden with lipoid. In the kidneys a few of the convoluted tubules showed various degrees of fatty degeneration. Dense groups of round cells were present in a few areas. The heart, thyroid gland, lymph nodes, pancreas, bone marrow and stomach were normal.

Dog G.—Grossly all the organs were essentially normal.

The results of microscopic examination were as follows: The liver was moderately congested. The lungs showed moderate congestion of the alveolar capillaries. The islands of Langerhans were rather numerous. The kidneys were moderately congested. The heart, thyroid gland, lymph nodes, spleen, suprarenals, bone marrow, stomach, parathyroid glands, testicles and epididymis were normal.

*Comment.*—The gross and microscopic studies made did not indicate abnormal changes in the brains of animals acutely and chronically

poisoned by dinitrophenol, at least according to the methods used. There were, moreover, no detectable changes in the other organs and tissues which could be ascribed to the effects of the drug. Therefore, these results were consistent with the negative chemical findings described.

#### SUMMARY AND CONCLUSIONS

Six dogs receiving di-nitrophenol continuously during a period of six months were observed for possible deleterious effects, as indicated by examinations of the urine, blood and tissues. Three of the animals were given 5 mg. and three 10 mg. of 2, 4, di-nitrophenol per kilogram of body weight daily by mouth. Three dogs without medication were used as controls.

Estimations at intervals of three weeks of the amount of sugar and albumin in the urine and of the hemoglobin and red, white and differential blood cell counts, urea content, icteric index and oxygen capacity of the blood and fragility of the red cells showed no significant or consistent deviations from the normal or control values.

Examinations of the brains of these nine dogs and of three dogs killed with acutely fatal doses of di-nitrophenol showed no demonstrable pathologic changes.

Other organs and tissues of the nine dogs in the series given continuous medication showed no significant pathologic changes grossly or microscopically.

It therefore appears that doses of di-nitrophenol of over three times the usual clinical doses may be given to dogs continuously for at least six months without causing demonstrable injury of vital functions or organs.

## Notes and News

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**Evaluation of Serologic Methods for the Diagnosis of Syphilis.**—The Public Health Service, in cooperation with the American Society of Clinical Pathologists, has drafted a plan to evaluate the methods of serologic diagnosis of syphilis in this country.

**Support by the Rockefeller Foundation of Medical Sciences.**—In the medical sciences the Foundation, during 1933, appropriated \$1,173,853. In aid of programs of specific concentration in the fields of psychiatry and public health teaching, appropriations were made to the Johns Hopkins University School of Medicine, Baltimore, for research in psychiatry; to University College, London, for work in biophysics and neurophysiology; to Washington University, St. Louis, for investigations in nerve physiology, and to the Harvard University Medical School, Boston, and the Massachusetts General Hospital for cooperative work in psychiatry. For the development of teaching in public health and preventive medicine an appropriation was made to Dalhousie University, Halifax, Nova Scotia. During 1933 the Foundation provided two hundred and ninety-five fellowships in the medical sciences. In addition, research aid grants in sums varying from \$55 to \$3,000 enabled sixty-one scientists or groups of scientists to carry on research work.

**University News, Promotions, Resignations, Appointments, Deaths, etc.**—Frank L. Horsfall, John W. Murray and Albert B. Sabin have been appointed as assistants on the staff of the Rockefeller Institute for Medical Research.

Bailey K. Ashford, Colonel in the Medical Corps of the United States Army and widely known for his pioneer work on hookworm, died on November 1, at the age of 61.

**Society News.**—The next meeting of the Federation of American Societies for Experimental Biology, in which is included the American Society for Experimental Pathology, will be held in Detroit, April 10 to 13, 1935.

The annual Gross Lecture of the Pathological Society of Philadelphia was given on the evening of December 13 by Dr. Shields Warren, who spoke on "Recent Advances in the Pathology of the Thyroid Gland."

# Abstracts from Current Literature

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## Experimental Pathology and Pathologic Physiology

THE HEART VALVES AND MUSCLE IN EXPERIMENTAL SCURVY WITH INFECTION.  
JAMES F. RINEHART and S. R. METTIER, Am. J. Path. **10**:61, 1934.

On the basis of the experimental data reported, and other experimental, epidemiologic and clinical data previously outlined, the theory is advanced that a condition of vitamin C undernutrition may be a necessary background for the development of rheumatic fever; when the insult of infection is combined with the scorbutic state, the pathologic picture of rheumatic fever develops.

THE DEVELOPMENT OF THE PULMONARY SILICOTIC NODULE IN THE EXPERIMENTAL ANIMAL. W. S. LEMON and G. M. HIGGINS, Am. Rev. Tuberc. **28**:470, 1933.

The authors present a new method of introducing suspensions of silica into the lungs of rabbits (using sodium amyta for anesthesia) by means of a tracheal cannula and roentgen localization. With this method they were able to follow the various changes that produce the typical nodule—that build it up, encapsulate and obliterate it, or that disintegrate it, destroy and eliminate it. Such progressive steps cannot be witnessed in the study of silicosis in man. The first and youngest silicotic nodule became recognizable within forty-eight hours. It was a small spherical bit of consolidated parenchyma, lying eccentric to a small bronchus. Within from three to five days strands of fibrous tissue were formed by the fibroblasts in the periphery of the nodule. The lesion became surrounded by them, and they penetrated into the central portions where silica was most abundant. Within seven days the first typical pulmonary silicotic nodules were completed. Within the second week necrosis was well advanced in the central zone, and between the first and sixth months organization was complete. The nodule either became completely fibrotic, or, after first becoming necrotic, showed replacement of the necrosis by mononuclear cells of the histiocytic type which produced argyrophil fibers in vast numbers and obliterated the nodule. Within that same period, however, reactions of degeneration were also observed, in which small or large abscesses had developed. Erosion into the bronchi or the blood vessels or the pleural spaces occurred, and death often resulted from subsequent pneumonia, sepsis or empyema. The foregoing metamorphosis of the typical pulmonary silicotic lesion was completed within six months.

H. J. CORPER.

LEAD IN MULTIPLE SCLEROSIS. W. CONE, C. RUSSEL and R. U. HARWOOD, Arch. Neurol. & Psychiat. **31**:236, 1934.

Lead was found in the urine, spinal fluid and feces in multiple sclerosis. The fact is emphasized that lead may be absorbed early in life and retained by the bones. Under certain conditions (pregnancy, state of acidosis, lactation and diet poor in calcium) it may be liberated, flood the system and produce changes typical of multiple sclerosis.

GEORGE B. HASSIN.

CEREBRAL FAT EMBOLISM. L. S. MERIWETHER and D. C. WILSON, Arch. Neurol. & Psychiat. **31**:338, 1934.

The reaction of microglia in cerebral fat embolism was studied in twenty grown rabbits which received, through the carotid artery, 8 minims (0.48 cc.) of olive oil stained with scarlet red. The animals were kept alive for from two hours to thirty days. The injected fat escaped through the capillary walls and as early as four hours after the injection it was found within the microglial cells. Around

the areas of necrosis the astrocytes became numerous, their processes interlacing with proliferated connective tissue from the neighboring blood vessels and partly with the pial prolongations. An early cerebral change was edema on the side of the injection, but there was no hemorrhage.

GEORGE B. HASSIN.

ETIOLOGY OF AGRANULOCYTIC ANGINA. F. W. MADISON and T. L. SQUIER, J. A. M. A. **102**:755, 1934.

Evidence is presented that agranulocytic angina may depend to some degree on the use of drugs containing amidopyrine and barbiturates.

HEMOGLOBIN INJECTIONS IN RABBITS ANEMIC FROM BLEEDING. D. K. MILLER and C. P. RHOADS, J. Exper. Med. **59**:333, 1934.

An anemia characterized by an increase in the size of the red blood cells and a high color index was produced in rabbits by repeated bleeding and by subcutaneous injection of stroma-free hemoglobin solution. The bone marrow of these rabbits reverted to a more primitive stage than did the marrow of rabbits rendered anemic in the same manner but not treated with hemoglobin.

FROM THE AUTHORS' SUMMARY.

EXPERIMENTAL HYPERTENSION. H. GOLDBLATT et al., J. Exper. Med. **59**:347, 1934.

These experiments indicate that, in dogs at least, ischemia localized to the kidneys is a sufficient condition for the production of persistently elevated systolic blood pressure. When the constriction of both main renal arteries is made only moderately severe in the beginning, the elevation of systolic blood pressure is unaccompanied by signs of materially decreased renal function. In this respect the hypertension in these animals resembles that which is associated with so-called benign nephrosclerosis in man. Subsequent increase of the constriction of the main renal arteries does not materially damage renal function, probably because of an adequate development of accessory circulation. More delicate methods for detecting a change may yet prove that some damage occurs. Almost complete constriction of both main renal arteries from the beginning results in great elevation of systolic blood pressure, which is accompanied by severe disturbance of renal function and uremia. This resembles the type of hypertension which is associated with so-called malignant nephrosclerosis, in the sense of Fahr. In several of the animals with persistent elevation of systolic blood pressure, anatomic changes were observed in the glomeruli, vessels and parenchyma of the kidneys which are most probably directly referable to the ischemia.

FROM THE AUTHORS' CONCLUSIONS.

EFFECT OF EXTRACT OF THE ANTERIOR PITUITARY ON THE EARLY GROWTH OF THE ALBINO RAT. A. M. TARGOW, J. Exper. Med. **59**:699, 1934.

Castrated male albino rats were given injections of a growth-promoting extract of the anterior lobe of the bovine pituitary gland from the twenty-first day of life (day of castration) to the fifty-sixth day (average for the group), at which time the difference in weight between the treated animals and the controls was first clearly discernible. In comparison with the controls the pituitary-treated animals at this stage showed: (1) a decrease in the weight of the pituitary gland; (2) an increase in the nose-anus length; (3) an increase in the weights of heart, lungs, liver and kidneys, which increase is shown to be of the nature of a splanchnomegaly; (4) an increase in the weight of the blood removed; (5) an increase in the water content of the skin and of the kidneys; (6) a tendency toward an increase in weight of the carcass.

FROM THE AUTHOR'S SUMMARY.

PORPHYRINS IN HUMAN DISEASE. V. R. MASON, CYRIL COURVILLE and E. ZISKIND, Medicine 12:355, 1933.

Three types of the disease may be recognized: congenital, acute toxic and acute idiopathic hematoporphyria. The congenital type is associated with porphyrinuria, staining of the bones and teeth and striking dermal photosensitivity (hydroa aestivale) appearing usually in the early years of life. The acute type usually appears first after twenty years of life and is characterized by porphyrinuria, acute, painful abdominal symptoms resembling ileus and severe degenerative lesions of the nervous system resembling Landry's paralysis. The majority of patients in this group have used barbituric acid compounds to excess, but in the remainder no causal factor can be discovered. The porphyrins in the urine are coproporphyrin and uroporphyrin. They are probably formed in the hematopoietic organ owing to some fault in the enzymatic synthesis of hemoglobin. The changes in the nervous system consist of extreme degeneration of the motor ganglion cells of the cord and the cerebellar Purkinje cells with slight alteration of the cortical cells. There are also degenerative changes in the posterior ganglion cells and in the abdominal sympathetic ganglion cells. The peripheral nerves show degeneration of both the myelin sheaths and the axis-cylinders. Round cell infiltration was found about the blood vessels and scattered through the neural bundles in the sciatic nerves. Yellow pigment was present in the perivascular spaces of the brain and choroid plexus.

AUTHORS' SUMMARY.

EXPERIMENTAL TOXIC APPROACH TO MENTAL DISEASE. A. FERRARO and J. E. KILMAN, Psychiat. Quart. 7:115, 1933.

The authors studied the intoxications induced by indol, histamine and potassium cyanide. When cats were given varying amounts of indol subcutaneously, from which all of the animals eventually died, the brain showed varying degrees of degeneration of the cells of the cerebrum, thalamus and hypothalamus as well as slight degenerative changes in the astrocytes and oligodendroglia. Subcutaneous injections of varying amounts of histamine caused slight degenerative changes of the Nissl cells and a central fatty infiltration of the liver, with increase in the lymphoid tissue of the mucosa and submucosa of the intestines. Subcutaneous injections of indol and histamine caused marked lesions in the central nervous system. There were diffuse cerebral lesions, mostly ischemic foci with Nissl degeneration. The neuroglia, astrocytes and oligodendroglia were also degenerated. The liver showed a severe fatty degeneration. In cats given injections of indol and potassium cyanide, the brain changes were similar to but less marked than those caused by indol and histamine combined. It is suggested that small doses of indol, histamine or potassium cyanide may be highly injurious to the central nervous system. The hypothesis is advanced that mental disease may result from a systemic absorption of such poisons from the digestive tract.

WILLIAM FREEMAN.

TRANSPLANTATION OF LIVER. G. R. CAMERON and C. L. OAKLEY, J. Path. & Bact. 38:17, 1934.

In an autoplasic liver transplant the peripheral cells survive and give rise to new bile ducts and liver cells. Proliferation of the surviving cells commences within two days, each type of cell growing independently and showing no tendency to unite with the other. The liver cells are, however, unstable and soon undergo degeneration and atrophy, with the formation of pigment. The bile ducts persist for long periods unchanged except for dilatation and flattening of their lining cells. Since the grafts are well vascularized it is suggested that the failure of the liver cells to maintain their existence is due to some inherent difference from duct cells in their make-up. Homoiplastic and heteroplastic transplants undergo complete destruction and are eventually absorbed.

AUTHORS' SUMMARY.

**EXPERIMENTAL PNEUMOCONIOSIS: INFECTIVE SILICATOSIS.** E. H. KETTLE, J. Path. & Bact. **38:**201, 1934.

These experiments form part of a wider investigation which is still in progress, but in themselves they appear to be sufficiently advanced to throw light on two of the fundamental problems in the pathology of pneumoconiosis, namely, the nature of the dust causing the disease, and the nature of the lesions provoked by it.

*Nature of the Dust.*—In many industries the working conditions entail the production of dusts, which are suspended in the atmosphere and are harmful to those who inhale them, since they are capable of producing extensive pulmonary fibrosis. In some industries these dusts are relatively simple in their composition; in others they consist of a variety of substances; but in all the occupations in which there is a recognizable dust hazard a considerable proportion of the suspended particles consists of some form of silicon. In fact, according to modern views, pneumoconiosis and silicosis are practically synonymous terms. It is not generally accepted, however, that all forms of silicon are productive of silicosis. The oxide, silica, is universally acknowledged to be dangerous; but with the single exception of asbestos, a complex silicate of iron and magnesium, it is commonly held that silicates are innocuous. Since there is general agreement that quartz and other varieties of crystalline silica are not harmful when inhaled into the lung, merely because of their physical state, there seems little reason why other compounds of silicon should not also possess the power of producing pulmonary fibrosis, and, indeed, W. R. Jones in 1933 brought forward a strong case in support of the view that sericite, a potassium aluminum silicate, is the cause of much of the industrial silicosis. For some time I have been working with an allied silicate, kaolin, and have shown that it possesses the characteristics of a potentially dangerous dust. The present experiments show that kaolin is capable under certain conditions of causing extensive lesions when introduced into the lungs of guinea-pigs and is, indeed, one of the most active dusts with which I have worked.

*Nature of the Lesions.*—It is generally acknowledged that even a serious degree of pulmonary fibrosis may be caused by the inhalation of silica without any accessory factor, but this pure or simple silicosis is rarely of clinical significance; nearly always silicosis is associated with tuberculosis, and my experience with silicosis in this country leads me to place increasing importance on the infective factor. I rarely see a case of simple silicosis, and even in those cases in which tuberculosis appeared to have been definitely grafted on to a silicotic lung, there has often seemed to me to be strong evidence that the apparently pneumoconiotic lesions were infective from the beginning. These experiments support this view, for they show that lesions can be rapidly produced in the lungs of guinea-pigs when a dust is combined with an infective process, whereas lesions can be produced only with the greatest difficulty, if at all, by the dust alone.

## FROM THE AUTHOR'S CONCLUSIONS.

**THE MANIFOLD EFFECTS OF CASTRATION IN MALE RATS.** V. KORENCHEVSKY and M. DENNISON, J. Path. & Bact. **38:**231, 1934.

Castration in male rats produces the following definite changes: striking atrophy of the sex organs (prostate, seminal vesicles and penis), slight atrophy of the thyroid gland and, about seventy days after castration, of the liver and kidneys; delay in involution of the thymus and hypertrophy of the suprarenal glands and hypophysis. In addition most animals show an increase in the deposition of fat and a decrease in body weight or in gain of body weight of small and varying degree. It is possible that the very slight decrease in the weight of the heart, which, on the average, was observed in most castrated rats as compared with their normal litter mates may also be specific. These changes are produced by changes in the tissues of the organs and not by changes in the blood content.

## FROM THE AUTHORS' SUMMARY.

FAT MOBILIZATION IN STARVATION. J. HENRY DIBLE and J. LIBMAN, J. Path. & Bact. **39**:269, 1934.

Our earlier finding that in the rat the extent of the fat infiltration in the liver in starvation is determined by the quantity of fat available in the animal's storage depots has been confirmed in rabbits starved for periods up to seven days and losing as much as 27 per cent of body weight. An increased percentage of fat in the liver cell is a constant result of starvation, but in animals whose stored fat is meager the increase may be only a relative one, since in these animals the shrinkage of the liver in starvation is early and marked. On the other hand where a sufficient supply of mobilizable fat is available there is an absolute increase in hepatic fat. In very adipose individuals in the earlier stages of starvation this may more than counterbalance any loss in the weight of the liver.

FROM THE AUTHORS' SUMMARY.

XEROPHTHALMIA, TRIGEMINAL DEGENERATION AND VITAMIN A DEFICIENCY. E. MELLANBY, J. Path. & Bact. **38**:391, 1934.

Evidence is given which suggests that xerophthalmia produced in animals by diets deficient in vitamin A and carotene may be secondary to a loss of the neurotrophic control normally exerted on the cornea by the ophthalmic division of the trigeminal nerve. When xerophthalmia is present, the corresponding trigeminal nerve usually shows degenerative changes in the myelin sheaths, and in the rabbit their development is commonly almost synchronous. In early and slight xerophthalmia when the corneal epithelium becomes normal as the result of adding carotene or vitamin A to the diet, the nerve also becomes normal. In such cases the myelin degeneration is of the annular type, and although the fibers may be swollen the myelin does not invade the axis-cylinder. This type of nerve degeneration seems capable of rapid recovery. In more severe cases of xerophthalmia, in addition to degeneration of the annular type, many of the fibers show typical wallerian degeneration, the axis-cylinder being invaded by degenerated myelin, while globules of the latter are scattered throughout the nerve. In cases of this type recovery of the nerve fiber either does not take place or does so only after months of vitamin A or carotene therapy. The nerve cells of the gasserian ganglion as well as their nerve fibers show degenerative changes. Indeed it is possible that the original lesion is in the cells, and that the trigeminal changes, peripheral and central, are secondary thereto. In the rat, in which as the result of vitamin A deficiency the eyelids become puffy, the cornea may escape xerophthalmic changes. In such cases also the trigeminal nerve shows demyelination. Since degeneration of the afferent nerves is widespread in animals brought up under these experimental conditions, it is probable that hyperplasia and metaplasia of other epithelial and mucosal surfaces throughout the body, and the subsequent invasion of these tissues by micro-organisms, are also related to changes in their afferent nerve supply.

FROM THE AUTHOR'S SUMMARY.

OEDEMA IN EXPERIMENTAL NEPHRITIS. J. S. DUNN, E. G. OASTLER and S. L. THOMPSETT, J. Path. & Bact. **38**:421, 1934.

Extensive subcutaneous edema and serous transudates can be obtained in nephritis produced by mercuric chloride and potassium dichromate as well as by uranium, provided the renal lesions are severe enough and the animals ingest sufficient fluid. The greater liability to edema in uranium nephritis is probably due to the severe damage to renal structure and function which uranium causes, combined with low general toxicity and noninterference with nutrition. The essential cause of the edema in experimental tubular nephritis is failure of the kidney to excrete sufficient fluid. Oliguria is due to failure of the damaged renal tubules as conducting channels.

FROM THE AUTHORS' CONCLUSIONS.

RESEARCH ON BURNS. S. NICOLAU and P. POINCLOUX, Ann. Inst. Pasteur 52: 217, 1934.

"In the blood of burned animals (rabbits, man) there develops a special leukocytic reaction characterized by: (a) multiplication of large mononuclears; (b) development of young primordial cells; (c) appearance of the cells of Rieder, and (d) slight increase in Türk cells. This reaction, which we call 'megamono-nucleosis,' is precocious, intense, lasting; it is proportional to the intensity and to the extent of the burns; it is interpreted, we believe, as a defense of the organism of the burned against the intoxication to which it is submitted. That those burned are submitted to an intoxication may be shown by intra-abdominal injection of their blood into a mouse (from 0.5 to 2 cc.), which often kills these animals. The principle toxic for mice is more abundant in the whole blood than in the serum; it exists in the liver in sufficient quantity to be demonstrated. Blood of those burned, or that of an animal which has succumbed to the injection of such blood, induces flocculation of ordinary meat broth. Anesthesia exerts a definite protective action against the risk of rapid death to which animals exposed to heat are subject."

#### AUTHORS' CONCLUSIONS.

EXPERIMENTAL INTESTINAL OBSTRUCTION. J. BOTTIN, Arch. internat. de. méd. expér. 9:51, 1934.

In dogs subjected to intestinal obstruction at a point 5 or 6 cm. below the mouth of the main duct of the pancreas, a concentration of blood elements with a relative diminution in blood volume developed, with a progressive fall in arterial blood pressure. Bottin concludes that the syndrome in high intestinal obstruction is due largely to dehydration.

RALPH FULLER.

FORMATION OF BONE AND CARTILAGE AFTER INJECTIONS OF QUININE HYDRO-CHLORIDE. R. SEVERI, Pathologica 25:611, 1933.

Rabbits were given intramuscular injections of a solution of quinine hydrochloride. Formation of bone in the muscle resulted in two animals, and of cartilage in three animals. Experiments on dogs and white mice failed because the salt is toxic for these animals.

E. VON HAAM.

NORMAL AND PATHOLOGIC HISTOPHYSIOLOGY OF THE HUMAN THYROID EPITHELIUM. JOHANNES WAHLBERG, Arb. a. d. path. Inst. Univ. Helsingfors 7:197, 1933.

In the normal thyroid the greater part of the parenchyma is inactive and serves as a functional reserve. Colloid is stored in most of the follicles. This occurs through a process of secretion in the cytoplasm in the form of vacuoles, which are emptied into the follicular cavity. In a certain number of follicles, the hormone follicles, there is a reversal of the process of secretion from the apical to the basal portion of the cell, where large vacuoles are formed which empty directly into the capillaries or the intercellular spaces and lymphatics. In colloid goiter the apical direction of secretion is pathologically increased but is qualitatively similar to the normal process. In thyrotoxicosis the histophysiology indicates that there is an excessive secretion of thyroid hormone. In cases of goiter without clinical signs of toxicosis in which mechanical pressure is relieved by surgical means, with subsequent relief of nervous and circulatory symptoms not due to pressure, microscopic examination shows definite evidence of thyrotoxicosis. The clinical improvement after preoperative treatment with iodine in thyrotoxicosis corresponds to the histophysiologic reversal of polarity in secretion from basal to apical type with storage of colloid in the follicles.

JACOB KLEIN.

ARTIFICIAL ENDOMETRIOSIS. H. H. SCHMID, Arch. f. Gynäk. **155**:217, 1933.

Since 1929 Schmid has practiced vaginal transplantation of uterine mucosa where hysterectomy had to be done. The purpose was to encourage menstruation for its psychic and physical benefits. The procedure, which is quite simple and harmless, consists in the insertion of some uterine mucosa between the posterior vaginal wall and the peritoneum. Thus far there have been nineteen cases in which such transplantation was successful. The autotransplants healed and maintained normal menstrual function.

JACOB KLEIN.

## Pathologic Anatomy

SWELLING OF THE MICROGLIA: REACTION TO INTOXICATION. ELI MARCOVITZ AND BERNARD J. ALPERS, Arch. Neurol. & Psychiat. **31**:1045, 1934.

While the behavior of the microglia in destructive lesions of the central nervous system has been well studied, its behavior in toxic conditions is much less well understood. From experiments and studies on human material Marcovitz and Alpers have come to the conclusion that microglia shows definite changes in acute and chronic states. They injected intravenously and in one case subdurally phosphorated oil (N. F.) two or three times a week into seventeen male rabbits, and killed them at intervals of from a few days to several weeks later. The changes in acute cases were loss of spines and side branches and swelling of the process, nucleus and cytoplasm. Later the processes became thicker and broke up into smaller fragments, and the cell body became vacuolated. In subacute and chronic cases (many weeks' duration) the cells including the spines became hypertrophied but finally exhibited varicosities and vacuolation of the branches. No lipoids were found in them, nor were ganglion cell changes present. The authors found similar changes in human brains in Alzheimer's disease, poliomyelitis, meningitis and meningo-encephalitis. The lesion is mainly regressive, although it possesses some reparative features.

GEORGE B. HASSIN.

HISTOLOGY OF THE CENTRAL NERVOUS SYSTEM IN THE ST. LOUIS EPIDEMIC OF ENCEPHALITIS. ARTHUR WEIL, Arch. Neurol. & Psychiat. **31**:1139, 1934.

As epidemic encephalitis may present a great variety of clinical pictures it does not seem reasonable to consider the slight variations presented by the St. Louis epidemic (occurrence in the summer months, absence of oculomotor nerve paralysis, restlessness preceding somnolence) as signs of a new type of encephalitis. Nor have the histologic changes differed from those seen in the classic type, as shown by the observations of Weil on eight brains from patients who died during the 1933 epidemic of encephalitis in St. Louis. He found a mild leptomeningitis, especially marked at the base of the brain; perivascular infiltrations of the veins and arteries, mainly by lymphocytes; formation of glia nodules in the white substance, in which oligodendroglia predominated; absence of hemorrhages; mild ganglion cell changes, and a predominance of inflammatory changes in the thalamencephalon and midbrain. In two cases, similar inflammatory phenomena were also present in the spinal cord. Micro-organisms (in the form of diplococci, short chains of cocci or small agglutinated colonies) were found in the subarachnoid space or within the walls of smaller blood vessels and occasionally within foci of inflammatory reaction.

GEORGE B. HASSIN.

ENCEPHALOMALACIA IN INFANTS. I. B. DIAMOND, Arch. Neurol. & Psychiat. **31**: 1153, 1934.

Cerebral changes in infants described by Virchow as interstitial encephalitis have been studied by Diamond in two infants, aged 7 weeks and 3½ months, respectively. He found marked degenerative changes with formation of lacunae, vast accumulations of fat granule bodies, proliferation of blood vessels, reduction

in size of the affected hemisphere, reactive glial phenomena and so-called aseptic meningitis (accumulation in the subarachnoid space of proliferated fibroblasts, mesothelial cells, polyblasts, lymphocytes and gitter cells). The changes were considered by Virchow to be inflammatory and a manifestation of fatty metamorphosis of the glia, while later observers held trauma to be the most common, if not the only, cause. In Diamond's cases the cause of the changes justly classified as degenerative was in one case a marked infectious process which started in an eye and necessitated its enucleation, and, in the second case, a severe intoxication.

GEORGE B. HASSIN.

**SCLEROTIC ATROPHY OF THE CEREBELLUM.** GEORGE B. HASSIN, Arch. Neurol. & Psychiat. **31:**1205, 1934.

In two cases of sclerotic atrophy of the cerebellum, a condition known under many other names (e. g., "primary parenchymatous atrophy" or "late cerebellar cortical atrophy"), there was practically a complete destruction of the ganglion cells of the molecular, Purkinje cell and granular layers with secondary involvement of the white substance including the mossy and climbing fibers. The destroyed parenchyma was replaced by a feltwork of glial fibers and a row of so-called Bergmann's glia nuclei. In exceptionally rare instances, Purkinje cells could be discerned, but they were calcified and misplaced. The degeneration was in patches and was confined to a few leaflets of the semilunar and a part of the quadrilateral lobe. As the patches resulted from primary destruction of the gray matter (the ganglion cells) and secondary involvement of the white substance, they differed from those of multiple sclerosis, in which not the ganglion cells but the white matter is primarily destroyed. In spite of such a far-gone cerebellar lesion clinical cerebellar signs were not in evidence. The clinical picture in one case was leukemia; in the other, dementia praecox.

AUTHOR'S ABSTRACT.

**BIPOLAR SPONGIOBLASTOMA IN THE REGION OF THE HYPOTHALAMUS, ASSOCIATED WITH INFANTILISM AND WITHOUT DWARFISM.** WALLACE B. HAMBY, Arch. Neurol. & Psychiat. **31:**1258, 1934.

Infantilism caused not by a pituitary lesion but by a glioma of the hypothalamic region was observed by Hamby in a man aged 22, who six months prior to examination began to suffer from headaches, vomiting and later from rapidly failing vision. For two years there were polydipsia and polyuria. The patient was of large size, his sexual organs and distribution of fat were infantile, and there was lack of axillary hair. There was a marked bilateral papilledema with a binasal hemianopia, and the roentgen examination revealed unfused epiphyses at the lower ends of the radii and ulnae and the bones of the hands and a calcified supracellar tumor. At necropsy the tumor proved to be a bipolar spongioblastoma. It was located above the hypophysis, almost filled the entire third ventricle and consisted of spindle-shaped cells, rich in cytoplasm, which "streamed away from the nucleus in two long processes" and contained a fairly large nucleus. Some cells were multinucleated.

GEORGE B. HASSIN.

**A CONSTITUTIONAL ABNORMALITY OF THE POLYMORPH LEUCOCYTES.** A. F. B. SHAW, J. Path. & Bact. **38:**259, 1934.

A case is described of a persistent deflection of the polymorphonuclear count to the left associated with an abnormally complex nuclear configuration of all the class I cells. There is no nuclear change in the other leukocytes, and the total and differential counts are normal. It is claimed that the condition is a constitutional abnormality of the polymorphonuclear leukocytes. Hereditary transmission was not found, but the evidence is incomplete. No record of a similar case has been found in the literature, and the condition differs in certain important respects from "Pelger's familial anomaly of nuclei." FROM THE AUTHOR'S SUMMARY.

INFLAMMATION IN THE CATERPILLARS OF LEPIDOPTERA. G. R. CAMERON, J. Path. & Bact. **38**:441, 1934.

Caterpillars possess an effective phagocytic mechanism composed of (1) the blood cells, of which there are three main types: (a) lymphocytes, (b) leukocytes and (c) spherule cells; (2) the pericardial cells; (3) certain cells in the fat body.

DISSECTING ANEURYSMS. T. SHENNAN, Medical Research Council, Spec. Rep. Ser. no. 193, 1934.

This report gives an analysis of 273 cases of dissecting aneurysm described in the literature and 17 cases studied by the author himself. It concerns chiefly dissecting aneurysms of the aorta. The feature common to all the cases is degeneration in the media, which usually gives way before the intima ruptures. This degeneration usually arises "as a result of toxic insults of varying character sustained during life." Syphilis does not play a prominent part in the genesis of dissecting aneurysm. Proximity of the primary tear to the heart appears to favor slightly the extent of the dissection and to shorten the period of survival. The dissection distal to the primary rupture is usually more important and more extensive than that proximal to the rupture. Shennan regards the predominance of primary ruptures in the ascending aorta as explainable by the abrupt diastolic recoil meeting the resistance of the closed aortic valve. The distal reentrance of the dissecting blood into the lumen, which allows free circulation through the new channel is "the most important determining factor in the healing process."

FROM THE AUTHOR'S SUMMARY.

HISTOLOGIC LESIONS IN EXPERIMENTAL PNEUMONIC PLAGUE. J. BABLET and G. GIRARD, Ann. Inst. Pasteur **52**:155, 1934.

Guinea-pigs were intratracheally inoculated. Alveolar foci with invasion of the pulmonary lymphatic system were established and multiplied within forty-eight hours, without breaking into the blood capillaries. Experimental pneumonic plague was thus definitely established. The evolution of the infection was histologically similar to that of human plague.

FROM THE AUTHORS' CONCLUSIONS.

THE ENDOCRINE GLANDS IN CIRRHOSIS OF THE LIVER. C. FITTIPALDI, Pathologica **25**:248, 1933.

In an anatomic study of the endocrine glands in the course of cirrhosis of the liver, Fittipaldi found that the parathyroid and pineal glands showed histologic changes in only a few cases. In the hypophysis and the suprarenal glands very slight changes were noted. The thyroid gland, pancreas and testicles, however, showed degenerative changes in every case. In most of these cases the changes may be considered as senile or arteriosclerotic processes. He believes, therefore, that there is no correlation between the endocrine glands and the cirrhotic process, except that the changes in the glands may be considered to be an expression of an abnormal condition, in the sense of an imbalance of the mesenchyma.

E. VON HAAM.

FATTY INFARCT OF THE LIVER. S. MARRAS, Pathologica **25**:798, 1933.

Two cases of fatty infarct of the liver (Cesaris Demel, A.: *Pathologica* **24**:532, 1932) are described. The condition is a fatty degeneration in the form of a sharply limited infarct-like focus, easily recognized by its yellow color and present without diffuse degeneration. It arises from a circumscribed degeneration of small branches of the hepatic artery with deficient oxygenation of the corresponding hepatic tissue.

E. VON HAAM.

NATURE AND ORIGIN OF HYALINE THROMBI OF THE CEREBRAL VESSELS. M. MANDELSTAMM, Beitr. z. path. Anat. u. z. allg. Path. **92**:476, 1934.

A series of more than a hundred brains were examined microscopically. Hyaline thrombi were observed in the cerebral capillaries and postcapillaries somewhat more frequently in cases of infection and in certain psychoses, but bore no very definite relation to the general condition before death. The thrombi are not composed of fibrin or cellular derivatives, but consist of a lipoid protein combination that is coagulated out of the blood plasma during a state of capillary stasis during which water is withdrawn from the vessels into the surrounding brain tissue. The globular or beaded shape of the masses is brought about by capillary contraction acting on the viscid coagulum.

O. T. SCHULTZ.

ERYTHROLEUKOBLASTOSIS, ICTERUS GRAVIS AND CONGENITAL ANASARCA. R. PETERS, Beitr. z. path. Anat. u. z. allg. Path. **92**:531, 1934.

Peters presents a study of a case of icterus gravis and one of congenital anasarca in children of the same mother. The infant with anasarca, a boy who died immediately after birth, was born two and one-half years after the infant, a girl, who died on the third day after birth of icterus gravis. Two previous children were normal, as were both parents. Common to both the children who died was marked erythroleukoblastosis in the liver, spleen and kidneys. The author tabulates and discusses seventy-five cases of congenital anasarca and fifty-two of icterus gravis in which there was microscopic examination. With Schridde, von Gierke and others, he considers both conditions to be manifestations of the same congenital maldevelopment, the outstanding characteristic of which is erythroleukoblastosis. Icterus gravis is frequently associated with ascites, and congenital anasarca with jaundice. Congenital anasarca is held to be the more severe grade of the process. The infants are usually born prematurely and are dead at birth or die shortly thereafter. In icterus gravis the duration of pregnancy is longer and may be normal; the child lives for a few days, and some probably recover.

O. T. SCHULTZ.

SIZE AND NUCLEAR, LEUKOCYTIC AND FAT CONTENT OF THE RENAL GLOMERULUS. O. SOMMER, Beitr. z. path. Anat. u. z. allg. Path. **92**:567, 1934.

In the kidneys obtained at 100 consecutive, unselected necropsies embracing all age periods up to the eighty-first year, the absolute weight, the weight in relation to body weight, the size of the glomeruli, and their nuclear, leukocytic and lipoid contents were determined. The size was determined by measuring the maximum and minimum diameters of 30 glomeruli in each kidney; the nuclei were counted in an equal number. The object of the investigation was to establish norms for various age periods. The weight of the kidney increases progressively and rapidly from 25 Gm. at birth to 260 Gm. at 20 years, remaining fairly unknown during the rest of life. The weight of the kidney relative to body weight undergoes a marked and rapid increase immediately after birth, ascribed to the rapid development of glomeruli and their taking on of function at birth; the relative weight then drops rapidly, to maintain a fairly even level throughout life. The average diameter of the glomeruli increases from 80 microns at birth to 175 microns at 30 years and remains fairly uniform until 60 years, when a slight progressive decrease occurs. The number of glomerular nuclei increases from 120 at birth to 155 at 35 years, beyond which period there is a slight decrease. The average number of oxidase-positive leukocytes increases from 4 at birth to 8 at 20 years and then remains fairly uniform; the number appears to depend on the blood content of the glomerulus. Stainable lipoid in the capsular epithelium increases rapidly from the fifteenth to the thirteenth year and then more slowly. The material is held to be a wear-and-tear pigment.

O. T. SCHULTZ.

**BILATERAL RENAL APLASIA.** H. MADESSON, Centralbl. f. allg. Path. u. path. Anat. **60:1**, 1934.

Following a review of the embryology of the kidney are detailed reports of necropsies on four new-born children in whose bodies no kidneys were found. All four of these children were males weighing from 1,500 to 3,200 Gm. who were born after long labors, up to sixty-six hours. In three bodies there were no kidneys, ureters or renal arteries; in one the left ureter coursed from a fibrous nodule in the region of the left suprarenal gland to an uncertain termination in the pelvis of the body. In the latter there was no urinary bladder, and only a lumenless fold of skin was present at the normal site of the penis. A penis, patent distally for from 5 to 15 mm., was present on each of the other three bodies. Two of these babies were born of the same parents, a normal sister being born in the interval between the two boys. The children exhibiting these marked anomalies lived from twenty to sixty minutes and apparently died of asphyxia. The author reviews the theories of the causation of renal aplasia and believes that the conditions which he encountered can best be explained on a chromosome deficiency basis.

GEORGE RUKSTINAT.

**MYOCARDIAL CHANGES IN EPILEPSY AND THEIR RELATIONS TO ANGINA PECTORIS.** K. NEUBÜRGER, Frankfurt. Ztschr. f. Path. **46:14**, 1933.

Gruber and Lanz reported changes in the myocardium in epilepsy. Neubürger examined the hearts of thirty-four patients who suffered from epilepsy and who died before reaching the age of 40. In fourteen he found myocardial changes characterized by simple degenerations of the myocardial fibers, progressive necrosis and scars. The changes were more common in the left ventricle, particularly in the cranial portions of the papillary muscles. An interference with the nutrition of the myocardium from spasm of the smaller vessels is held responsible. It is also possible that tonic contractions of the heart over a long period of time may have led to a mechanical compression of small branches of the coronary arteries, resulting in vascular disturbances. The author believes that coincidental with every severe epileptic attack there may also be an attack of angina pectoris. The latter might escape clinical observation because of the severity of the epileptic fit. Death occurring during the epileptic attack in some instances is the result of the myocardial changes which, however, may be difficult to demonstrate histologically.

O. SAPHIR.

**CONGENITAL ADENOMYOSIS.** R. WEYENETH, Frankfurt. Ztschr. f. Path. **46:43**, 1933.

In forty-two girls aged up to 14 years four instances of endometrial structures within the myometrium were found. Various other congenital anomalies were present also, including a case of true hermaphroditism, which is described in detail. The author also describes a case of uterus didelphys with endometrial structures throughout the entire left half. Adenomyosis of the uterus and of the tube should be regarded as malformations resulting from extension of the endometrium into the myometrium during the fetal period or from cutting off of epithelial elements of the müllerian duct.

O. SAPHIR.

**THE PARATHYROID GLANDS IN DISEASES OF THE KIDNEYS.** P. RADNAI, Frankfurt. Ztschr. f. Path. **46:97**, 1933.

There is disturbance in the calcium metabolism in chronic nephritis and nephrosclerosis, and recently changes have been described in the parathyroid glands in disturbed calcium metabolism. For these reasons Radnai examined the parathyroids in primary renal disease. He concludes that there are relatively more acidophilic cells in these glands in renal disease than otherwise.

O. SAPHIR.

THE NATURE AND HISTOGENESIS OF SO-CALLED BASAL CELL CARCINOMA.  
M. GLASUNOW, Frankfurt. *Ztschr. f. Path.* **46**:140, 1933.

Basal cell carcinoma is characterized by slow course, superficially destructive growth, absence of metastasis and cachexia, and marked sensitivity to chemical and physical factors. Only ectodermal regions are involved. In most cases the tumor occurs in the skin of the face. Up to date it has not been possible to produce this type of tumor experimentally. Because of the various histologic forms of the tumor it seems likely that it arises from a primary multipotent cell of embryonal character. On the face the tumor often arises in the region of the point of union of the facial clefts.

O. SAPHIR.

SEVERE NECROSIS OF THE COLON FROM CONTUSION OF THE ABDOMEN. F. WAGNER, Frankfurt. *Ztschr. f. Path.* **46**:185, 1933.

A 50 year old man suffered a severe trauma of the abdomen. Four days later severe abdominal pains set in with fever. The patient died two days after operation. At autopsy intestinal perforation could not be found. In many places the muscularis of the colon was torn, and here the mucosa and submucosa projected into the peritoneal cavity. The tears in the muscularis were the direct result of the trauma. Formation of thrombi within the vessels of the intestines secondarily led to the necrosis of the mucosa and submucosa.

O. SAPHIR.

CHANGES IN THE LIVER IN THYROTOXICOSIS. R. RÖSSLER, *Virchows Arch. f. path. Anat.* **291**:1, 1933.

A double number of *Virchows Archiv*, comprising 570 pages, consists of contributions from Rössler's institute in Berlin. In the opening article Rössler describes and discusses changes in the liver in thyrotoxicosis which he has seen repeatedly in the course of the past twenty years but which have received little attention in the literature. The gross alterations are slight; interlobular fibrosis of the subcapsular zone is sometimes evident to the naked eye. The histologic changes in selected cases are described as illustrative of the five groups into which Rössler divided his material: cases with old changes; cases with slight and slowly developing alterations; cases with recurrent changes; cases with acute alterations, and those with an unusual course ending in cirrhosis. The acute alterations consist in necroses in the central zone or about the central vein of the lobules; the process may be so marked as to simulate acute or subacute yellow atrophy. Associated with the necroses are focal changes in the capillary endothelium leading to altered distribution of the blood and to serous exudation, which leads Rössler to use the term "serous hepatitis." In later stages sclerosis of the parenchyma occurs, especially in the subcapsular zone; this process may be associated with true cirrhosis. No single alteration is characteristic of thyrotoxicosis, but the series of changes is characteristic and is due to the thyrotoxicosis. The sclerosis is due to fibrils that are formed independently of fibril-forming cells, that is, fibroblasts and capillary endothelium. The fibrils are formed by physicochemical action in the serous exudate as in a culture medium. Rössler believes that such acellular fibril formation may be an important factor in the sclerosis of other organs.

O. T. SCHULTZ.

MALIGNANT NEPHROSCLEROSIS. P. SCHÜRMANN AND H. E. MACMAHON, *Virchows Arch. f. path. Anat.* **291**:47, 1933.

In a profusely illustrated monograph of 172 pages, the authors discuss the clinical symptomatology and the gross and microscopic pathology of malignant nephrosclerosis based on a very full and detailed description of thirty-seven cases. This is followed by a section devoted to the differential diagnosis of malignant nephrosclerosis from the benign form, and from glomerulonephritis, periarteritis nodosa and other cardiovascular-renal diseases. In a final section they discuss the

pathogenesis and nature of the essential vascular lesion of malignant nephrosclerosis. This lesion is arteriolonecrosis, in contrast to the arteriosclerosis of benign nephrosclerosis. The necrosis is the result of degeneration or injury of the endothelium and its basement membrane. The endothelium is looked on as a barrier that normally protects the blood and the subendothelial tissues of the arteriole and of the organ from each other. When this barrier is broken the blood acts harmfully on the subendothelial tissues, causing hyperplasia, histolysis or necrosis, depending on the degree of action. The tissues and tissue fluids in turn act harmfully on the blood, causing precipitation of fibrin. In the arterioles and renal glomerular vessels the precipitation of fibrin leads to thrombosis and necrosis. For this process of destruction of the blood-tissue barrier the word "dysoria" has been coined, the root of the word coming from the Greek word meaning barrier. Two forms of malignant nephrosclerosis are recognized. One begins with arterial hypertension, which may be the result of a variety of factors, and leads to dysoria and the full-blown picture of malignant nephrosclerosis; this is the genuine form of Volhard. The other form begins acutely as a primary renal disease resulting from exogenous toxic factors and proceeding to the characteristic vascular lesion; this is the exogenous toxic form of Fahr.

O. T. SCHULTZ.

ATYPICAL AMYLOIDOSIS. A. STRAUSS, Virchows Arch. f. path. Anat. **291**:218, 1933.

The literature contains records of a gradually increasing number of examples of amyloidosis that differ from the usual, typical form in the absence of involvement of the organs characteristically involved in true amyloidosis, in a predominant involvement of the walls of the smaller muscular arteries and of striped muscle and adipose tissue, in the absence of an underlying condition like suppuration to which the condition may be due, and in the atypical character of the microchemical reactions. For this atypical form of amyloidosis the name "paramyloidosis" is proposed. The condition reported by Strauss occurred in a man aged 72. Deposition of material morphologically like amyloid but not giving all the characteristic reactions of this substance had increased the thickness of the walls of smaller arteries up to ten times the normal. On the epicardium and pericardium was a layer of similar material from 1 to 2 cm. thick. It was present widely in adipose tissue. Necrosis and chronic suppuration were absent. Strauss briefly discusses twenty-eight previously reported cases of atypical amyloidosis. Common to all was the rather advanced age of the persons with paramyloidosis.

O. T. SCHULTZ.

MEASUREMENT OF THE CONSISTENCY OF THE BRAIN. R. NEUMANN, Virchows Arch. f. path. Anat. **291**:341, 1933.

The sclerometer and Brinell methods of determining consistency were found to be not applicable to the brain because of the semifluid character of this organ. Neumann devised and fully describes an apparatus that measures consistency by the depth to which a cone of definite weight, size and shape penetrates the tissue by its own weight in the space of five seconds. The results are recorded in the form of an enlarged graph on a revolving kymograph drum. Differences in penetration of 0.1 mm. are accurately recorded. The mathematical analysis of the graphs, which looks very complicated to an old-fashioned pathologist, is described in detail. With this apparatus the consistency of 100 brains, removed as soon after death as possible, was determined, 100 to 200 determinations being made on each brain. The cerebrum was cut into a series of frontal sections, the pons and medulla into transverse sections, and the lateral lobes of the cerebellum into sagittal sections. On these tissue slabs the consistency of the various white and gray areas was determined. The consistency increased during the first thirty to forty hours after death and then decreased. Changes in consistency resulting from

a variety of factors affected chiefly the white matter. The consistency of hyperemic brains was higher than that of anemic brains. The consistency of edematous brains varied from high to low. Swelling of the brain, especially in the presence of acute infection, was associated with diminished consistency. The consistency of atrophic brains was increased. There was no correlation between consistency of the brain and sex, age and habitus. In diabetes mellitus the brain had an unusually high consistency, described as a peculiar density different from that of the sclerotic brain, which could not be accounted for by histologic alterations. In disturbed fat metabolism the consistency was also increased. The brain in a case of systemic paramyloidosis had a high consistency, although microscopic examination did not reveal the presence of amyloid.

O. T. SCHULTZ.

**DETERMINATION OF THE MUSCLE MASS OF THE STOMACH.** K. HELMKE, *Virchows Arch. f. path. Anat.* **291:**507, 1933.

One hundred and forty-four human stomachs were used in the study here reported. The stomach was opened, laid flat, and its surface area determined by measurement. All external extraneous tissue was removed down to the peritoneum. The mucosa and submucosa were removed together from the fibromuscular coats. The weight of the muscle mass and that of the mucosa and submucosa were determined in the fresh, moist state and after drying to constant weight. The fat contents of the two portions were determined. The average weight of the muscle mass of the normal stomach was 0.87 Gm. per kilogram of body weight; that of the mucosa and submucosa was 1.2 Gm. The maximum weight of the muscle mass was 3.1 Gm. per kilogram of body weight, in a man with pyloric stenosis due to ulcer. The minimum weight was 0.57 Gm., in a man with pulmonary tuberculosis. The weight of the muscle was increased in pyloric obstruction, diabetes mellitus and hepatic cirrhosis. The mucosa and submucosa made up 58 per cent of the total weight of the stomach. This proportion was decreased in achylia, pernicious anemia, old age and chronic gastritis. It was increased in plethora and in some cases of chronic gastric catarrh. The water content of the wall of the stomach was increased in uremic gastritis, passive congestion, ascites, cirrhosis of the liver and sepsis. The fat content of the wall paralleled that of the body. The mucosa of the diabetic stomach contained exceptionally large amounts of fat. The average surface area of the normal stomach was 473 sq. cm. The maximum observed was 792 sq. cm., in the stomach with the highest muscle weight. Small fibromyomas at the cardia were seen in 10 per cent of the stomachs examined.

O. T. SCHULTZ.

**HYPERTROPHY OF THE GUMS.** F. GERLING, *Virchows Arch. f. path. Anat.* **291:** 522, 1933.

Gerling presents two cases of hypertrophy of the gums. In the first, that of a girl who died at the age of 10 years, the condition had been present since the fifth year of life. Previous to this time the child had had encephalitis followed by hemiplegia. In psychoses in which the mouth is held open, hypertrophy of the gums has been noted. The child began to menstruate at 7 years of age and exhibited other evidences of precocious puberty, supposedly the result of endocrine disturbance. In previously reported cases gingival hypertrophy has been ascribed to endocrine dysfunction. Gerling thinks that both factors may have played a part in this case. The enlargement of the gums was due to diffuse increase in collagenous connective tissue, which did not appear to be inflammatory in origin. The differentiation of this condition from hyperplastic gingivitis is discussed. The second case was one of acute myeloblastic leukemia. The gingival hypertrophy was due to myeloblastic infiltration of the tissues of the gums.

O. T. SCHULTZ.

A CASE OF CLEIDOCRANIAL DYSOSTOSIS. P. LADEWIG, *Virchows Arch. f. path. Anat.* **291**:542, 1933.

The rare condition that P. Marie and Sainton described in 1898 under the name "dysostosis cleidocranialis hereditaria" is characterized by small body stature; by maldevelopment of the cranial vault with prominence of the frontal and parietal regions; by late or deficient ossification of sutures and fontanelles, which are closed by multiple irregular masses and nodules of bone; by an abnormally small face; by deformity of the jaws with many unerupted teeth, and by bilateral partial or complete absence of the clavicles. There may be other skeletal deformities. Anatomic descriptions are meager. This Ladewig ascribes to the fact that the persons with the deformities are usually well psychically and as regards their general health; the condition goes unrecognized and escapes the pathologist. In the case described by Ladewig, that of a woman who died at the age of 60 years of carcinoma of the uterus, the body had gone first to the anatomic institute, where it had been partially dissected before its peculiarities were recognized. His description is therefore limited chiefly to the skull. He concludes that the condition is a congenital and probably hereditary one, characterized by diminished capacity of expansion on the part of the ossification centers, especially those of the membranous bones. Similar but less extremely diminished growth energy is evident in the rest of the skeleton.

O. T. SCHULTZ.

### Microbiology and Parasitology

THE POSTMORTEM REMOVAL OF LIVER TISSUE FROM RAPIDLY FATAL FEVER CASES FOR THE DISCOVERY OF SILENT YELLOW FEVER FOCI. F. L. SOPER, E. R. RICKARD and P. J. CRAWFORD, *Am. J. Hyg.* **19**:549, 1934.

The organization of a service for the routine postmortem collection of tissues from the liver in cases of rapidly fatal fever resulted, between May 1930 and June 1933, in the examination of specimens from over twenty-eight thousand livers, and a pathologic diagnosis of yellow fever in fifty-four cases from forty-three places in which yellow fever was not known to be present. A special instrument designed for the rapid removal of hepatic tissue by laymen without autopsy, to which the name "viscerotome" has been given, is described. The routine collection and examination of liver has resulted in: (a) the demonstration of typical fatal cases of yellow fever in foci in which otherwise the disease is silently endemic; (b) the accumulation of extensive data indicating, so far as negative evidence can indicate, the absence of the yellow fever virus from key centers of population and from hundreds of towns and villages in which antimosquito measures are being applied; (c) the confirmation of the occurrence of yellow fever without *Aedes aegypti* in a small village eleven months after the occurrence of the first suspected case; (d) the accumulation of valuable data regarding the distribution of diseases other than yellow fever producing characteristic lesions of the liver.

FROM THE AUTHORS' SUMMARY AND CONCLUSIONS.

THE MICROSCOPICAL EXAMINATION OF 29,593 HUMAN LIVERS FROM CENTRAL AND NORTHERN BRAZIL WITH SPECIAL REFERENCE TO THE OCCURRENCE OF MALARIA AND SCHISTOSOMIASIS. N. C. DAVIS, *Am. J. Hyg.* **19**:567, 1934.

There have been examined 29,593 specimens of livers from persons dying in central or northern Brazil between May 1, 1930 and June 30, 1933. Most specimens were secured with the viscerotome (described by Soper, Rickard and Crawford: *Am. J. Hyg.* **19**:549, 1934). Malarial pigmentation was recognized in 2,298 specimens. The highest rates of malarial infection were found in Pará, Amazonas and Bahia, in the order given. The lowest rate was in Ceará. The months of greatest incidence were May and June. The age group of highest absolute incidence was that of from 15 to 19 years, although the differences between this group and the two preceding groups (from 5 to 9 and from 10 to 14 years)

were not significant. Lesions caused by *Schistosoma mansoni* Sambon were recognized in 1,594 livers. The highest rates of schistosome infection were found in Sergipe, Alagoas, Pernambuco and Bahia, in this order. Appreciable rates were found in Parahyba, Espírito Santo and Rio Grande do Norte. In other states the rates were almost negligible. The seasonal fluctuations was less noticeable than those of malaria. The age group of highest incidence was again that of from 15 to 19 years, although the rates did not differ significantly in the groups between the ages of 10 and 29 years. Central necrosis was unduly prevalent in livers with malarial pigmentation and especially so in those with schistosome granulomas. The geographic and seasonal distribution of other cases in which central necrosis was noted suggests that malaria (including blackwater fever) and schistosomiasis may have been responsible for an appreciable proportion of the lesions, even though pigmentation or granulomas were not detected. Lesions of enteric fever and focal necroses of doubtful etiology (many probably typhoid) were especially prevalent among specimens from Ceará. It is explained that this finding was undoubtedly correlated with unsanitary conditions fostered by a severe drought in that state. The rate for miliary tuberculosis was high in the Amazon basin and low in Ceará, Parahyba and Rio Grande do Norte. Various grades of periportal inflammation, with fibrosis shading into marked cirrhosis, were very common.

FROM THE AUTHOR'S SUMMARY.

**THE DISTRIBUTION AND EPIDEMIOLOGY OF HUMAN ASCARIASIS IN THE UNITED STATES.** G. F. OTTO and W. W. CORT, Am. J. Hyg. **19**:657, 1934.

The evidence presented from our own and other surveys gives a rather clear picture of the distribution of Ascaris in the United States. No endemic centers of importance to public health are recorded for the northern and western parts of the country. In the southeastern states the problem is found among the indigenous population of the Appalachian mountains and the foothills running, south, east and west from the mountains. Certain other centers of infestation are found in south-central Louisiana, in southeastern North Carolina and in the city of Tampa, Fla. For the most part, however, the lowland and costal plain areas of the southern United States have a very low incidence of ascariasis. This rather peculiar distribution of Ascaris in the southern United States can be correlated with the presence or absence of factors favorable to its continued dissemination. These factors have been described at some length and may be summarized under two heads: (1) habits favorable to pollution and (2) climatic conditions.

FROM THE AUTHORS' SUMMARY.

**SARCOSPORIDIA IN THE MYOCARDIUM OF A PREMATURE INFANT.** A. T. HERTIG, Am. J. Path. **10**:413, 1934.

A case of sarcosporidiosis involving the myocardium of a 26 day old premature infant is reported. This was an incidental observation in the routine microscopic study of postmortem material. The mode of entrance of the parasites into the body cannot be stated with certainty.

FROM THE AUTHOR'S SUMMARY.

**EXPERIMENTAL TUBERCULOSIS OF THE BONES AND JOINTS OF RABBITS.** PAUL J. TRUDEL, Am. Rev. Tuberc. **28**:331, 1933.

The experimental production of tuberculosis in the bones and joints of rabbits requires delicate technic, but such technic yields good results in 85 per cent of the animals. While only few of the nonsensitized rabbits given injections of human bacilli revealed small tuberculous foci in the lungs and liver, 75 per cent of all the animals first sensitized to and then given infective injections of bovine bacilli showed striking localization in the bones and joints with no detectable focus elsewhere. Only small doses (from 0.05 to 0.15 mg.) should be injected. The bacillary emulsion should be injected into the nutrient artery or the nutrient foramen of

the femur. If the size of the artery does not permit injection, a direct inoculation of the bacilli into the synovial membrane of the knee may be substituted. The bovine type of bacillus gave far better and quicker results than the human type.

H. J. CORPER.

**THE VIABILITY AND VIRULENCE OF OLD CULTURES OF TUBERCLE BACILLI.**  
H. J. CORPER and MAURICE L. COHN, Am. Rev. Tuberc. 28:856, 1933.

In 1919 and 1920 several hundred nursing bottles containing about 3 ounces (90 cc.) of 5 per cent glycerol broth, neutral to litmus, were planted with nineteen different strains of human and bovine tubercle bacilli and were maintained at 37 C. until 1932, when forty-seven bottles containing human bacilli were chosen to determine the viability of the bacilli, and twenty were found to contain bacilli capable of growing on egg yolk medium; of nine bottles planted with bovine bacilli, four contained cultures that were viable in 1932. Twelve of the original seventeen human strains were recovered for study while, of two bovine strains, one was recovered. No growth occurred in transplants from bottles in which the  $p_H$  was below 6.1 or above 7.6. About 1 per cent of the bovine bacilli and 0.01 per cent of the human bacilli in the culture masses tested were viable. The virulent strains had not suffered a diminution of virulence during the twelve years' residence in the incubator, and strains of low virulence remained of low virulence during this time. Virulence is a property unchangeable in individual bacilli, but prolonged cultivation on artificial mediums may cause a perceptible, though not striking, change in the progeny of human tubercle bacilli. Persistence of bacilli in the body is dependent on a number of factors, but prominent among these, aside from the pathogenicity of the bacilli, is the factor of the number of tubercle bacilli introduced into the organs and tissues of an animal.

H. J. CORPER.

**THE INHIBITORY EFFECT OF NORMAL BLOOD ON THE GROWTH OF TUBERCLE BACILLI.** H. J. CORPER and C. B. VIDAL, Am. Rev. Tuberc. 28:878, 1933.

The growth of human and bovine tubercle bacilli on good mediums is inhibited by normal blood from dogs or rabbits. This inhibition is more evident with small numbers than with large numbers of bacilli and is more evident on inspissated egg yolk than on potato. The effect is due to the development of toxic autolytic products from normal blood, and is absent when the blood has been treated with one or two volumes of 6 per cent sulphuric acid for from one-half to one hour at 37 C., which destroys the autolytic enzymes, followed by neutralization with isotonic sodium bicarbonate solution (or other nontoxic alkalies). Regardless of the treatment, however, accurate determination of the number of tubercle bacilli placed in blood is not possible by counting the colonies which this blood will yield on good mediums suited to growing small numbers of human and bovine tubercle bacilli from the suspensions.

H. J. CORPER.

**THE EFFECT OF ANIMAL PASSAGE ON THE VIRULENCE OF TUBERCLE BACILLI.**  
HENRY STUART WILLIS, Am. Rev. Tuberc. 28:884, 1933.

An account is given of the passage of a strain of tubercle bacilli (RI) of low virulence through guinea-pigs for a period of ten years. This required fifty-seven passages. During this prolonged period progressive diseases appeared three times, in three of about nine hundred animals used. Whether the disease in these instances was an extraneous, contaminating tuberculosis could not be determined. That it was is possible if not probable. But for these three exceptions passage appeared to have no effect on the virulence of the strain used. A strain of virulent tubercle bacilli was carried in animals for more than eight years through fifty passages with evidence of a slight increase of virulence. This strain carried on artificial mediums during the same eight-year period lost considerably in virulence

but is still fully capable, in small doses, of leading to the death of guinea-pigs. It appears that a prolonged stay of weakly virulent tubercle bacilli in animal tissues does not as a rule affect the virulence.

H. J. CORPER.

**PATHOLOGICAL FINDINGS AND PATHOGENESIS OF CONGENITAL TUBERCULOSIS.**  
MORRIS SIEGAL, Am. Rev. Tuberc. **29**:297, 1934.

Siegal reports on the findings recorded in thirty-eight cases of undoubted or probable congenital tuberculosis. The tuberculous organs were usually the lungs, liver and spleen and, after a certain age, the intestines. Macroscopically the lesions almost always resembled tubercles. Microscopically they were reported as tubercles, except those in the lungs, where tuberculous pneumonic areas were not uncommon. Caseation was common; cavitation, ulceration and calcification were also mentioned. In the few cases in which primary lesions were specifically reported, they were described as localized caseous areas in the lungs or the liver and in one instance as a diffuse bilateral tuberculous bronchiolitis and alveolitis with beginning caseation. The lungs were most frequently involved. The degree of pulmonary involvement was usually equal to and occasionally greater than that of the liver. In cases in which death occurred in the latter half of the first month of life, the lungs and regional lymph nodes were usually alone involved. Involvement of the liver and spleen went side by side, these organs usually being involved in the same cases and to the same degree. The liver was most frequently involved and to the greatest degree in cases in which death occurred in the first two weeks of life, least involved in those in which death took place in the second half-month of life, and usually involved in cases in which death occurred subsequently to this. Tuberculous lesions in the gastro-intestinal tract were seen in 70 per cent of infants of 39 days or older, all of whom had advanced pulmonary tuberculosis. No undoubted cases were reported in infants less than 39 days of age. The lymphatic nodes usually involved were the pulmonary, mesenteric and portal in diminishing frequency. Acute tuberculous leptomeningitis was reported in only one case.

H. J. CORPER.

**TUBERCLE BACILLI IN THE GASTRIC CONTENTS OF TUBERCULOUS CHILDREN.**  
INA GOURLEY, Am. Rev. Tuberc. **29**:461, 1934.

Fifty-nine cases of tuberculosis in childhood were studied by gastric lavage and the inoculation of guinea-pigs. All the children reacted positively to the intracutaneous injection of tuberculin. In fifty cases lesions of the lungs were demonstrable in roentgenograms. In nine there were no demonstrable lesions. In twenty-eight cases (47.7 per cent) tubercle bacilli were found in the gastric contents. Of the fifty children with demonstrable lesions, twenty-eight (56 per cent) had the bacilli in the gastric contents. Fourteen, or 50 per cent, of the children with the bacilli in the gastric contents were of school age. The study apparently indicates that in tuberculosis of childhood all types of parenchymal lesions, including those of the "C" group of McPhedran's classification (McPhedran, F. M.: Classification of Tuberculous Pulmonary Lesions in Childhood and Adolescence, Am. Rev. Tuberc. **20**:532, 1929), are "open."

H. J. CORPER.

**EFFECTS OF TUBERCULO-PROTEIN (MA-100) ON THE COURSE OF TUBERCULOSIS IN RABBITS AND GUINEA PIGS.** J. C. SMITHBURN, F. R. SABIN and J. T. GEIGER, Am. Rev. Tuberc. **29**:562, 1934.

Both rabbits and guinea-pigs, when subjected to a long series of daily injections of tuberculoprotein, became hypersensitive to this substance, as indicated by their intracutaneous reactions. Accompanying this induced hypersensitivity there was an abolition of the response in temperature induced by the protein. Little or no effect on resistance to tuberculosis in either rabbits or guinea-pigs was produced by the tuberculoprotein.

H. J. CORPER.

UNDULANT FEVER DUE TO BRUCELLA SUIS. C. P. BEATTIE and R. M. RICE,  
J. A. M. A. 102:1670, 1934.

In a milk-borne outbreak of undulant fever thirty cases occurred. Twenty-seven of the patients obtained their milk from the same dairy. The dairy, from a herd of twenty cows, supplied approximately eighty households; in eighteen of these, cases of undulant fever developed. *Brucella suis* was obtained in blood culture from six of fourteen patients and from the milk of one of the cows in the herd. The outbreak ceased thirteen days after the stoppage of the sale of milk from the dairy. There is a greater virulence of *Brucella suis* than of *Brucella abortus*. The possibility of milk containing *Brucella suis* must be considered.

FROM THE AUTHORS' SUMMARY.

RAT-BITE FEVER ACQUIRED FROM A DOG. H. S. RIPLEY and H. M. VAN SANT,  
J. A. M. A. 102:1917, 1934.

*Spirillum morsus-muris* has been isolated from two cases of rat-bite fever. The animal vector was apparently a dog. The disease has been produced in dogs. The Kahn reaction was strongly positive in both patients and in the dog that survived, while the Kolmer-Wassermann reaction was usually negative or weakly positive. A patient with dementia paralytica was inoculated with the spirillum, and a typical lesion and symptoms of rat-bite fever developed. Flagella were demonstrated only with Burri's india ink method. The mouse proved to be a better diagnostic animal than the guinea-pig. In one case, infection was contracted when no abrasion of the skin was noted, suggesting ease of penetration by the organism. The clinical course of the disease showed marked variation. A course of from three to six treatments with arsphenamine seems advisable.

FROM THE AUTHORS' CONCLUSIONS.

THE VARIANT AND FILTERABLE FORMS OF CERTAIN GREEN-PRODUCING STREPTOCOCCI. R. A. MCKINNEY, J. Bact. 27:373, 1934.

Rapid serial transfers of rough or smooth green-producing streptococci on a single medium induce a change in colony type but no change in morphology. Streptococci aged in unfavorable or in stimulating mediums and then subjected to repeated transfers on mediums of differing physical and chemical states produce variant colonies containing morphologically variant cells. Pleomorphic green-producing streptococci give rise by described technics to three variants—(1) a blood-bleaching, nitrite-forming diplococcus in rounded, moderately rough colonies, (2) a diphtheroid in gray or yellow mucoid colonies, and (3) a proteolytic spindle-shaped rod in definitely rough colonies. The diplococcus and the diphtheroid possess antigenic structures and biochemical functions differing from each other and from those possessed by the original streptococcus. Diplococci and diphtheroids so formed give rise again to streptococci by a process which is preceded by and probably depends on a changed mode of cell division. The streptococcus thus recovered resembles, but is not identical with, the original streptococcus in serologic properties and biochemical action. A precipitate containing viable forms occasionally develops in the filtrates of streptococcal cultures. This occurs when atypical cell division, chain reduction and granule formation are clearly evident in the culture before filtration. The viable forms eventually by a suitable technic assume a visible form, in most cases that of a diplococcus similar to the one isolated from the same culture before filtration. In two cases diplococci from the filtrates were converted to streptococci with attributes similar to but not identical with those of the original strains of streptococci.

FROM THE AUTHOR'S CONCLUSIONS.

LEISHMANIA DONOVANI IN NASAL AND ORAL SECRETIONS OF KALA-AZAR.  
C. E. FORKNER and L. S. ZIA, J. Exper. Med. 59:491, 1934.

Smears from the nasal cavities of fifteen patients suffering from kala-azar have been examined, and in those from nine of the patients typical Leishman-Donovan

bodies have been found. Smears from the surface of the tonsil and from the saliva of one of these nine patients showed Leishmania. The tonsils of this patient, who died as the result of kala-azar and secondary infection, were shown at autopsy to be massively infected with Leishman-Donovan bodies. Leishmania in the nasal discharges of two patients was shown, by inoculation into susceptible animals, to be viable and capable of producing infection. Sufficient time has not elapsed to determine the viability of the organisms in the remaining cases. These experiments show for the first time that in a large proportion of patients with kala-azar there is available a rich source of infective material for direct transmission of the disease. Strong evidence is presented that one of the natural routes, perhaps the most important natural route, of transmission of kala-azar is that from person to person by way of the upper respiratory and alimentary tracts. In two normal human volunteers and numerous normal experimental animals Leishmania has been inoculated into the nasal and oral cavities with the nasal discharge, known to contain Leishmania, from patients with kala-azar. The results of these experiments will be reported at a subsequent date.

FROM THE AUTHORS' SUMMARY AND CONCLUSIONS.

### Immunology

**SKIN REACTION TO TUBERCULIN PROTEINS IN HODGKIN'S DISEASE.** P. E. STEINER, Arch. Int. Med. 54:11, 1934.

Tuberculin protein (Seibert) was used in intracutaneous tests on thirty-five patients with Hodgkin's disease and on thirty-eight controls with a variety of lymphoma. Tuberculin proteins prepared from both avian and human strains were used. No evidence of specific sensitization to the avian tuberculin protein was obtained. A marked desensitization (or absence of sensitization) was found to both proteins in the patients with Hodgkin's disease. This relative absence of "normal adult sensitization" was of diagnostic value, especially in the differential diagnosis of tuberculous adenitis. The interpretation is made that either (1) the process of Hodgkin's disease desensitizes its victims to these tuberculin proteins or (2) Hodgkin's disease usually occurs in persons in whom development of the normal sensitization to the tuberculin protein is impossible. It is difficult to conceive of either of these phenomena as occurring in a disease unrelated to tuberculosis.

FROM THE AUTHOR'S SUMMARY.

**PATHOLOGIC HISTOLOGY OF THE SHWARTZMAN PHENOMENON.** H. T. KARSNER AND A. R. MORITZ, J. Exper. Med. 60:37, 1934.

Histologic study of the reaction to an injection of the preparatory factor of the Shwartzman phenomenon shows this reaction to be an exudative inflammation, but furnishes no conclusive evidence as to whether or not it is conditioned by previous sensitization to substances contained in the bacterial filtrate. Histologic study of the local reaction which follows an intravenous injection of the reacting or provocative factor shows an exudative inflammation which differs from that due to the preparatory factor principally in the increased general severity and the marked damage to blood vessels. The increased severity is probably due to a concentration of the injurious agent at the site of inflammation determined by the preparatory local injection. The exudative phenomena caused by the preparatory local injection are edema and infiltration by polymorphonuclear leukocytes and large mononuclear cells. Vascular injury is morphologically demonstrable in only a few of the sections, notably those from periaricular structures. The intravenous injection determines an increase in cellular infiltration, necrosis of exudate, phagocytosis of cell débris, destruction of vascular walls and hemorrhage. Healing is due to granulation, organization and cicatrization.

FROM THE AUTHORS' CONCLUSIONS.

## CHEMO-IMMUNOLOGICAL STUDIES ON CONJUGATED CARBOHYDRATE-PROTEINS.

W. F. GOEBEL, F. H. BABERS and O. T. AVERY, *J. Exper. Med.* **60**:85, 1934.

The results of the present study not only confirm the view previously held that the immunologic specificity of carbohydrates is determined by their stereochemical configuration, but lend support to the further assumption that the introduction of a simple chemical group, such as the acetyl radical, endows a carbohydrate with a new and distinct specificity which is determined by the chemical nature of the group thus introduced. It has been previously pointed out that the differences in the specific behavior of the alpha- and beta-glucosides of glucose may be accounted for by differences in the stereochemical configuration of the carbon atom bearing the aglucon, and that the basis for the immunologic crossing may lie in the fact that the spatial configuration of the polar groups on the remaining five carbon atoms is identical. This explanation appears to be further supported by the results of the present study. For it can be seen from table 2 that when beta-gluco-test antigen, which normally reacts in alpha-gluco-globulin antiserum, is so altered that one of the polar groups (OH) of the five terminal carbon atoms of the carbohydrate radical is replaced by acetyl ( $\text{CH}_3\text{CO}$ ), the resulting antigen fails to react in alpha-gluco-globulin antiserum. Furthermore, alpha-gluco-test antigen, which normally reacts with beta-gluco-globulin antiserum, fails to react in the immune serum produced by immunization with acetyl beta-gluco-globulin. Similarly, the acetylated beta-glucoside, owing to the alteration in chemical constitution, fails to bind the antibody in alpha-gluco-globulin antiserum, and as a result permits both alpha- and beta-gluco-test antigens to react in their normal course. The beta-glucoside likewise fails to inhibit the reaction between the acetyl beta-gluco-test antigen and homologous immune serum. This difference in the serologic specificity of the antibodies induced by beta-gluco-antigens in the acetylated and unacetylated form can be attributed only to the known differences in the chemical structure of these two glucosides. The latter differ from one another in that the acetyl beta-glucoside is a derivative in which one of the polar groups (OH) of the carbohydrate has been altered by the introduction of an acetyl radical ( $\text{CH}_3\text{CO}$ ). A critical analysis of the results of these serologic tests again emphasized the fact that the presence of an acetyl group in a carbohydrate exerts a determining influence on the specificity of an antigen of which it forms a part. In conclusion it may be pointed out that the differences in serologic specificity exhibited by the acetylated and deacetylated polysaccharides of *Pneumococcus* type I are accurately paralleled by the purely synthetic system described.

## FROM THE AUTHORS' DISCUSSION.

## THE ANTIGENIC RELATIONSHIP BETWEEN PROTEUS X-19 AND TYPHUS RICKETTSIA.

M. R. CASTANEDA, *J. Exper. Med.* **60**:119, 1934.

A soluble specific substance was isolated from the rickettsiae of Mexican typhus which gave, with *Proteus* X 19 antiserum and human typhus serum, the same precipitation reactions as the polysaccharides extracted from *Proteus* OX 19. The soluble specific substance extracted from the rickettsiae and *Proteus* OX 19 is likely to be of a polysaccharide nature, as judged by the strong Molisch reactions obtained with such extracts, the heat stability and the negative protein reactions (biuret). Since, however, it still contains 7 per cent nitrogen, this is not certain. In the antigenic composition of *Proteus* X 19 and typhus rickettsiae there is a common soluble specific factor which is responsible for the Weil-Felix reaction.

## FROM THE AUTHOR'S SUMMARY AND CONCLUSIONS.

## THE A SUBSTANCE OR ACETYL POLYSACCHARIDE OF PNEUMOCOCCUS TYPE I. J. F.

ENDERS AND C-J. WU, *J. Exper. Med.* **60**:127, 1934.

Since the evidence obtained in the course of this study indicates that the A carbohydrate of *Pneumococcus* type I (Enders, J.: *J. Exper. Med.* **52**:235, 1930) and the acetyl polysaccharide of Avery and Goebel (*J. Exper. Med.* **58**:731, 1925)

represent the same chemical substance, it is suggested that the designation "A carbohydrate" or "A substance" be relinquished in favor of the more exactly descriptive term "acetyl polysaccharide."

VACCINATION OF MONKEYS AND LABORATORY WORKERS AGAINST PSITTACOSIS.  
T. M. RIVERS AND F. F. SCHWENTKER, *J. Exper. Med.* **60**:211, 1934.

Monkeys that have recovered from psittacosis pneumonia have an increased resistance to infection with the virus and possess neutralizing antibodies in their serum. Large amounts of the active virus can be introduced intravenously and intramuscularly into monkeys without the production of a serious infection such as pneumonia. Relatively small amounts of the virus introduced intratracheally into monkeys usually lead to psittacosis pneumonia. Monkeys vaccinated intramuscularly with the unattenuated virus have an increased resistance to the active agent and possess neutralizing antibodies in their serum. The intramuscular introduction of the active virus in moderate amounts into human beings is relatively harmless, and repeated inoculations lead to the appearance of neutralizing antibodies in the serum of the vaccinated persons.

FROM THE AUTHORS' SUMMARY.

THE IMMUNOLOGICAL CHARACTERISTICS OF THE POLIOCIDAL SUBSTANCE IN HUMAN SERUM. C. W. JUNGEBLUT, *J. Immunol.* **27**:17, 1934.

Quantitative titrations of pooled serums of normal human adults and of adults convalescent from poliomyelitis revealed no fundamental differences in virucidal titer in the case of blood groups A and B. Normal serums of blood group O, on the other hand, slightly exceeded in potency the corresponding convalescent serums. The temperatures which destroyed the virucidal principle in normal and convalescent serums of the same blood groups were practically identical. The thermostability of a given serum varied in direct proportion to its virucidal titer. The virucidal substance could not be removed from either normal or convalescent serum by contact with virus *in vitro*. In several instances, however, absorption was observed after contact with various red cells. Neutralization of the virus *in vitro* was obtained with certain samples of antidiphtheritic, antistreptococcal and antisnake serum from the horse. In contrast to this, a large number of other immune horse and rabbit serums—antibacterial, antitoxic and antiviral—failed to inactivate the virus. Immunization of monkeys with diphtheritic toxoid or a toxin-antitoxin mixture increased the resistance of nearly one half of the animals in various degrees to subsequent poliomyelitic infection. Virucidal substances were demonstrable in some of the serums of completely or partially protected monkeys.

FROM THE AUTHOR'S CONCLUSIONS.

INACTIVATION OF POLIOMYELITIS VIRUS AND OF DIPHTHERIA TOXIN BY ENDOCRINE PRINCIPLES. C. W. JUNGEBLUT, K. MEYER AND E. T. ENGLE, *J. Immunol.* **27**:43, 1934.

The virus of poliomyelitis was inactivated *in vitro* by certain biologic products of human and animal origin containing anterior pituitary-like principles and by the suprarenal cortical and medullary hormones. Diphtheritic toxin was inactivated *in vitro* by preparations of the urine of pregnancy and by suprarenal cortical hormone.

FROM THE AUTHORS' SUMMARY.

THE AUTONOMIC NERVOUS SYSTEM IN THE ANAPHYLACTIC SMOOTH MUSCLE CONTRACTION. C. V. SEASTONE JR. and A. ROSENBLUETH, *J. Immunol.* **27**:57, 1934.

From the results reported it is evident that the contraction of the nictitating membrane of the cat furnishes a convenient criterion of anaphylactic shock in that animal. Since the smooth muscle concerned in these experiments was totally denervated, it is evident that the contraction during shock may occur without the

mediation of autonomic nerve impulses. The effectors are then directly excited during shock as they are by suprarenin, histamine and other substances.

FROM THE AUTHORS' DISCUSSION AND CONCLUSIONS.

THE EFFECT OF RETICULO-ENDOTHELIAL CELL BLOCKADE UPON ANTIBODY FORMATION IN RABBITS. L. TUFT, *J. Immunol.* **27**:63, 1934.

In rabbits efficient and continued blockade of the cells of the reticulo-endothelial system by such inert particulate material as india ink or trypan blue prevents the formation of agglutinins against *Bacillus typhosus* and *Bacillus paratyphosus A* and *B* and of complement-fixing antibodies against *B. typhosus*, and thus supplies further evidence of the importance of this system in the formation of these antibodies. The effect is independent of the route of administration of the antigen, as long as the dose of the antigen is not too large to cause excess stimulation. While the amount of particulate matter necessary for efficient blockade may vary with the type of antigen used and its affinity for the reticulo-endothelial cells, nevertheless the principle of continued saturation of these cells throughout the course of the experiment to prevent regeneration of new cells is essential for satisfactory results.

FROM THE AUTHOR'S CONCLUSIONS.

ELECTRIC CHARGE OF BACTERIA SENSITIZED WITH PURIFIED AGGLUTININS. L. OLITZKII, *J. Immunol.* **27**:105, 1934.

The H-forms of *Bacillus proteus* show a higher velocity of migration to the anode than do the corresponding O-forms. Agglutination with pure H-agglutinin reduces the charge of the agglutinable H-forms, while the inagglutinable O-forms in the same culture are not affected. Agglutination with pure O-agglutinin reduces the charge of the O-forms, the H-forms of the bacteria remaining unaffected. Agglutination with serum containing both types of agglutinins reduces the charge of all the cells present in the suspension. Below  $p_H$  4 the H-antigen was destroyed and H-agglutination did not occur. The bacteria treated with purified H-agglutinin were not agglutinated and showed the same anodic migration as unsensitized bacteria. Amphoteric protein-like behavior of bacteria, that is, a positive charge below the iso-electric point, was observed only after treatment with large amounts of nonpurified antiserum. After treatment with purified O-agglutinin or after washing the sensitized bacteria free from serum, cathodic migration below  $p_H$  4 was not observed.

FROM THE AUTHOR'S CONCLUSIONS.

THE SENSITIZATION OF GUINEA PIGS TO POISON IVY. F. A. SIMON AND OTHERS, *J. Immunol.* **27**:113, 1934.

Guinea-pigs can be made sensitive to poison ivy by rubbing a 10 per cent extract in petrolatum into their skin or by painting the normal skin with a strong extract. When, however, the extract is painted on the active lesions produced by the virus of cowpox the resulting sensitiveness is less marked. The response of the skin has been injured. When the extract is injected intravenously or intraperitoneally no cutaneous sensitiveness results. When one area of skin is sensitized, all the skin is sensitized. The production of a cutaneous sensitiveness to poison ivy has been accomplished so far only by treatment of the skin itself. These experiments give further support to the theory that the sensitiveness of man to poison ivy is acquired by virtue of previous contact with the plant.

THE ANTIGENIC VALUE OF THE "FIXED" VIRUS OF RABIES INACTIVATED BY PHOTODYNAMIC ACTION. I. A. GALLOWAY, *Brit. J. Exper. Path.* **15**:97, 1934.

The fixed virus of rabies is sensitive to the photodynamic action of methylthionine chloride (methylene blue). Under the conditions of the experiments it was inactivated in collodion-membrane or sand and paper-pulp filtrates but not in unfiltered suspensions. The virus appears to be relatively more sensitive to the

photodynamic action of proflavine than to that of methylthionine chloride, at least in the presence of physiologically active cells or portions of cells from an infected animal. It was inactivated by irradiation when the former dye was employed even in unfiltered suspensions of fresh infected brain. The fixed virus inactivated by the photodynamic action of methylthionine chloride or proflavine conserves its antigenic potency, since about twenty-six of thirty-one rabbits—84 per cent—which had received more than one dose of such a vaccine survived an intramuscular test dose of fresh virus, while of sixteen unvaccinated control animals only one survived (percentage of survival, 6).

FROM THE AUTHOR'S SUMMARY.

IMMUNIZING FRACTIONS FROM BACT. AERTRYCKE. H. RAISTRICK AND W. W. C. TOPLEY, Brit. J. Exper. Path. **15**:113, 1934.

In summary, we think it has been demonstrated that fractions can be isolated from *Bacterium aertrycke*, and inferentially from other organisms, that contain the specific somatic polysaccharides in an antigenically active form in the absence of any intact protein. Such fractions are toxic, and are efficient immunizing agents, inducing an active antibacterial and antitoxic immunity associated with the production of specific precipitating and agglutinating antibodies. The distribution of toxicity among the various fractions runs closely parallel to the distribution of immunizing potency. Whether this indicates that the toxic and immunizing substances are identical, or whether we are dealing with two or more different substances that behave in the same way to the reagents we have employed, or with an association resulting from adsorption, are problems that await further solution. It need hardly be added that there would be no justification for the assumption that the toxic and immunizing substances present in our fractions represent the total toxic and immunizing potentialities of the bacterial cells. The fractions with which we have been concerned are certainly not chemically pure substances, and we do not think that the evidence at present available, although indicative of the presence of a phosphatide as well as a polysaccharide, allows any final conclusion to be reached as to the exact structure of the complete antigenic molecule of which the polysaccharide forms a part.

FROM THE AUTHORS' SUMMARY.

BLOOD-SUGAR CHANGES AND TOXIC EFFECTS PRODUCED IN RABBITS BY FRACTIONS FROM BACT. AERTRYCKE. M. E. DELAFIELD, Brit. J. Exper. Path. **15**:130, 1934.

Fractions containing polysaccharide components but no unaltered protein, derived from the bodies of *Bacterium aertrycke* by tryptic digestion followed by alcohol precipitation, are active in producing hyperglycemia and toxic effects in rabbits. The toxicity of the various fractions tested runs closely parallel to their ability to induce antibacterial and antitoxic immunity in mice. These active fractions are all precipitable within the range of from 50 to 68 per cent by weight of alcohol concentration. The fraction obtained by excess alcohol after removal of the 68 per cent fraction is inactive, as is also the alcohol-soluble material. Acid or alkali hydrolysis of the alcohol precipitate designated F68/68 destroys the power to produce hyperglycemia. Immunity to the chemical response can be developed by a series of injections of the alcohol fraction F68/68.

FROM THE AUTHOR'S SUMMARY.

THE IMMUNIZING ACTION OF EXTRACTS OF PNEUMOCOCCI (TYPES 1 AND 2). D. HARLEY, Brit. J. Exper. Path. **15**:161, 1934.

Extracts of pneumococci (types 1 and 2) made by the action of twentieth-normal hydrochloric acid at 60 C. for fifty minutes produce active type-specific immunity in mice and type-specific agglutinins and protective antibodies in rabbits. Heating the extracts to 100 C. at  $p_{H_2} 4-5$  does not impair their immunizing action for mice, but similar treatment at  $p_{H_2} 9-10$  inactivates them. The relationship of the active principles of these extracts to the immunizing antigen of the intact

pneumococcus and to the acetyl polysaccharide of type I pneumococci (Avery and Goebel: *J. Exper. Med.* **58**:731, 1925) is discussed.

FROM THE AUTHOR'S SUMMARY.

EXPERIMENTS WITH THE "O" ANTIGEN OF CLOSTRIDIUM OEDEMATIS MALIGNI (VIBRION SEPTIQUE). D. W. HENDERSON, *Brit. J. Exper. Path.* **15**:166, 1934.

Active immunity can be obtained against Clostridium oedematis-maligni by the use of pure "O" antigen. The immunity is type-specific. A common "O" antigen, however, is shared by strains in varying degree and is effective in producing minimal degrees of cross-infection. In immunized animals the reaction to infection is generally associated with a severe local perforating gangrene. The "O" antigen functioning in agglutination and complement fixation is directly responsible for the production of the protecting immune body. A classification of strains of *Clostridium oedematis-maligni* is suggested. The basis of the primary differentiation should be the "O" antigen relationship, and the "H" antigen content should form the basis of a secondary grouping.

FROM THE AUTHOR'S SUMMARY.

THE PROBLEM OF NATURAL RESISTANCE IN THE WHITE RAT. E. DE BALOGH, *Ann. d'anat. path.* **10**:65, 1933.

Having previously shown that human tubercle bacilli injected into normal tissues of the white rat produced no damage, whereas when injected into transplantable tumors of this animal they produced necrosis and ulceration, the author made similar experiments with BCG. The same necrosis and ulceration resulted. Tumor tissue thus appears to be a susceptible portal of entry for an infection against which the animal is normally resistant. The experiment was repeated with *Bacillus proteus*, *Bacillus pyocyaneus*, *Bacillus diphtheriae*, *Bacillus typhosus* and *Bacillus coli*, and the same results were obtained. The necrosis following these injections occurred much more rapidly and was more intense than spontaneous necrosis in such tumors. Occasionally following such an injection the tumor would undergo complete necrosis, slough out and heal completely. The poor resistance of tumor tissue is not due to absence of inflammatory reaction, because such substances as turpentine, glycerin, iodoform, etc., when injected produce marked inflammatory changes in the stroma. It is more logical to believe that tumor tissue approaches the anergic state of embryonic tissue. Thus tumor tissue resembles embryonic tissue not only in its histologic and biochemical characters, but also in immunologic characters.

PERRY J. MELNICK.

### Tumors

MYXOMA OF THE HEART VALVES. T. C. JALESKI, *Am. J. Path.* **10**:399, 1934.

Whether the myxomas are neoplasms or whether they represent a degenerative process in connective tissue growths is still open to question.

MICROSCOPIC METASTASES IN THE THYROID GLAND. C. O. RICE, *Am. J. Path.* **10**:407, 1934.

In eighty-nine necropsies in cases of malignant growth tumor cells were found in the thyroid gland in nine. In five of these nine cases there were macroscopic secondary nodules; in the remaining four cases the metastases were microscopic only.

PARASELLAR MENINGEAL FIBROBLASTOMA ARISING FROM THE SPHENOID RIDGE. B. J. ALPERS and R. A. GROFF, *Arch. Neurol. & Psychiat.* **31**: 713, 1934.

Aside from intrasellar and suprasellar tumors, Alpers and Groff recognize parasellar tumors. These tumors are situated near the sella turcica and arise from

the sphenoid ridge, bearing a relationship to the lesser or greater wings of the sphenoid bone. They are encapsulated, lobulated in appearance and adherent to the dura, which they may invade. They may become calcified, extend to the middle and the anterior cranial fossa and compress the neighboring structures (second or third nerve, the internal carotid artery, the middle and anterior cerebral vessels). The tumors of the lesser wing are easy to diagnose; those of the greater wing give an indefinite clinical picture. Six of the tumors described resembled more or less typical meningeal fibroblastoma. Three were less typical.

G. B. HASSIN.

**LYMPHOSARCOMA IN BONE.** L. F. CRAVER and M. M. COPELAND, Arch. Surg. 28:809, 1934.

One hundred and sixty-four patients with lymphosarcoma have been studied. Seventeen patients (10.4 per cent) were found to have involvement of the bones. The bones most frequently involved were those of the spine and pelvis. Pathologic fracture was observed five times. Collapse of the vertebrae was relatively rare. Signs of compression of the cord were noted in three patients. Two types of osseous changes were noted: osteolytic and osteoplastic. Osteolytic changes predominated. A combination of both was seen occasionally. Two routes of involvement of the bone were observed: (1) the hematogenous and (2) direct infiltration from contiguous diseased lymph nodes. The histologic studies showed all the specimens to be reticulum cell lymphosarcomas. The stomach was involved by lymphosarcoma in two cases; the ileum, in one case. All the patients except one had evidence of involvement of the lymph nodes prior to demonstrable osseous changes. In two patients a low grade lymphatic leukemia developed prior to death. Histologically the lymph nodes showed typical lymphosarcoma.

FROM THE AUTHORS' SUMMARY.

**PRIMARY OSTEOPGENIC SARCOMA OF THE THYROID GLAND.** A. C. BRODERS and J. DEJ. PEMBERTON, Surg., Gynec. & Obst. 58:100, 1934.

This rare tumor occurred in a man 71 years of age. It was firmly adherent to the trachea, larynx and blood vessels, and consisted of polymorphic sarcoma cells, osteoid tissue and fully differentiated bone.

W. C. HUNTER.

**PRIMARY CARCINOMA OF THE URETER.** W. W. SCOTT, Surg., Gynec. & Obst. 58:215, 1934.

Primary carcinoma of the ureter is a relatively rare disease, there being only sixty-one acceptable cases recorded in the literature. The most common type of tumor is the papillary carcinoma. The disease occurs with equal frequency in the fifth, sixth and seventh decades of life. The average age of the patients in this series was 55.7 years. The right ureter seemed to be involved slightly more frequently than the left, and the lower third of the ureter was involved in 57 per cent of the cases. Increased knowledge concerning the behavior of this disease, as well as the marked advances in the technic of urologic diagnosis, should result in earlier diagnosis.

FROM THE AUTHOR'S SUMMARY (W. C. HUNTER).

**THE PRODUCTION OF DIBENZANTHRACENE TUMORS IN PURE STRAIN MICE.** H. B. ANDERVONT, Pub. Health Rep. 49:620, 1934.

The results of the experiment confirm the findings of Burrows, Hieger and Kennaway in showing that the subcutaneous injection of a dibenzanthracene-lard solution induces sarcomas in mice. In addition, it has been shown that this solution induces tumors in pure strain mice in which, under normal conditions, spontaneous tumors do not develop. Thus it is shown that the genetic constitution of a pure strain of mice does not prevent the cells from becoming malignant when exposed to

this carcinogenic agent. Transmission experiments demonstrate that the induced tumors grow only in mice of the same strain in which they originated. In this respect they are similar to spontaneous tumors arising in pure strain mice.

FROM THE AUTHOR'S SUMMARY.

THE INFLUENCE OF ISCHAEMIA ON THE DEVELOPMENT OF TUMOURS. JOHN W. ORR, Brit. J. Exper. Path. 15:73, 1934.

Fibrous scar tissue was produced in the subcutaneous tissues of mice by the insertion of linen thread sutures, which were subsequently removed, and healing was allowed to take place. Care was taken to avoid direct injury to the epithelium. Tar applications induced tumors more rapidly in these mice than in controls, and histologic examination showed that the percentage of carcinoma at the twenty-first week was twice that in the controls. Local injection of vasoconstrictor drugs produced an acceleration in the induction of tumors, more marked with epinephrine hydrochloride than with ephedrine sulphate. Carcinoma was increased in mice treated with epinephrine, but not significantly in those treated with ephedrine, as compared with controls receiving injections of salt solution and nontreated controls, at the twenty-first week of tarring. Tumors appeared more rapidly than usual where tar was applied after discontinuance of a series of injections of ephedrine sulphate. The results are discussed in relation to other work and certain concomitant conditions in cases of human cancer, and the opinion is expressed that carcinogenic agents act on cells which have been deprived of a fully adequate vascular supply.

FROM THE AUTHORS' SUMMARY.

AN ATTEMPT TO PRODUCE IMMUNITY TO INDUCED TUMOURS IN MICE. F. C. PYBUS, Brit. J. Exper. Path. 15:89, 1934.

The development of tar tumors was neither prevented nor delayed in mice which had received various numbers of inoculations of mouse embryo skin. Accordingly, the injection of skin has no protective action against the genesis of an induced tumor. Inoculations of mouse embryo skin continued after the appearance of tar tumors had no curative effect, and the tumors did not regress. The numbers of metastases found in the control mice were so much greater than those found in the inoculated mice that it is suggested that the inoculations of skin, although useless for producing an immunity to induced tumors, do develop a resistance in the animal against the metastatic spread of malignant cells.

FROM THE AUTHORS' SUMMARY.

IMMUNITY IN ROUS FOWL SARCOMA AND ITS BEARING ON THE PROBLEM OF THE NATURE OF NORMAL AND CANCEROUS GROWTH. M. J. A. DES LIGNERIS, Publ. South African Inst. M. Research 6:1, 1934.

"Finally, coming back to the tumour which has been investigated in our experiments, the Rous sarcoma, we have adopted the view that the agent, being in an intermediate position between the 'living and the dead,' unites with some part of the protoplasm of a congenial normal cell, and that this combine constitutes a somewhat new, alienated, living protoplasm, capable of metabolizing, of increasing at the expense of the protoplasm. Finally, the whole cell is taken up by 'tumour protoplasm' and we have a tumour cell of the malignant type, the assumed behavior of which (antigenic character, leading to antibody production by the organism and to retaliatory measures of self-defence and aggression by the tumour cell) we have just outlined.

"It may be said that this conception of tumours, leading from the normal cell, by intermediate stages up to the highly malignant Rous sarcoma cell, is not proved, that is only a hypothesis. We do not deny that we cannot by means of chemical formulae or mathematical deductions prove our case. In fact, such possi-

bilities are very rare in biology; but it is thought that, as no known facts are in conflict with this hypothesis, and as we know of no other hypothesis which would allow of such an easy explanation of all growth processes, without exception, we are fully permitted to use it as a working hypothesis, not only for theoretical discussions, but for the building up of practical deductions which should form a reasonable approach to research work on the prophylaxis of cancer."

**ANILINE TUMORS IN THE INDUSTRIAL REGION OF BASEL.** A. MÜLLER, Schweiz. med. Wchnschr. **14**:951, 1933.

Thirty-six cases of carcinoma of the bladder have been observed by Müller in chemical workers since 1912. The ages of the patients were from 22 to 77 years. The chemicals to which the workers were exposed by inhalation were amines of the aniline series. The average time from the beginning of exposure to the recognizable development of the tumor was sixteen years; the minimum time was six years. In these cases the carcinoma arose in the floor or walls of the bladder.

JACOB KLEIN.

**"MYOBLASTOMA" OF THE DIAPHRAGM.** W. MÜLLER, Centralbl. f. allg. Path. u. path. Anat. **58**:353, 1933.

The growth reported occurred on the right leaf of the diaphragm, was 10 by 7 by 4 cm., attached by a stalk 4 cm. in diameter, so that it was freely movable in the pleural cavity, and entirely covered with pleura. Microscopically the mass was heterogeneous, but most portions contained several well defined cell types. Striated muscle in bundles surrounded by thin connective tissue was evident, as were also regions rich in round or spindle-shaped cells with large nuclei, some of which contained mitotic figures. Typical myoblasts were not seen, but the author believes that the tumor should be classified as a malignant myoblastoma or myoblastic sarcoma.

GEORGE RUKSTINAT.

### Medicolegal Pathology

**HUMAN THALLOTOXICOSIS.** J. C. MUNCH, J. A. M. A. **102**:1929, 1934.

An extensive search of the literature prior to January 1934 has been made to learn the extent of human poisoning from thallium compounds and the number of deaths resulting therefrom. In the course of industrial exposure, 12 persons have been poisoned, but none has died. During clinical use, 692 persons have been affected and 31 have died. Toxicologic literature records 53 human beings poisoned by thallium compounds, with 10 deaths. Following rodenticidal and entomologic use, 21 human beings have been poisoned and 5 have died. Reports have been found of 778 human beings poisoned with thallium compounds; 46 (6 per cent) died with thallotoxicosis.

FROM THE AUTHOR'S SUMMARY.

**SPONTANEOUS DISLOCATION OF THE ATLAS.** F. A. R. STAMMERS and PHILIP FRAZER, Lancet **2**:1203, 1933.

Forward dislocation of the atlas on the axis is not exceptionally rare. It does not necessarily prove fatal, though it is more likely to do so if the odontoid process remains intact. Though usually associated with trauma (and this sometimes quite trivial) it may be essentially spontaneous, occurring during the course of: (a) acute rheumatic fever in any of its clinical forms; (b) septicemia, and (c) chronic non-specific arthritis of the spine (spondylitis deformans). The hypothesis that the dislocation, when spontaneous, is a distention luxation is supported.

FROM THE AUTHORS' CONCLUSIONS.

THE RÔLE OF AURICULOCARDIOPULMONARY REFLEX IN DEATH FROM DROWNING. E. FROMMEL, J. de physiol. et de path. gén. **31**:327, 1933.

Water under pressure in the external auditory canal may cause inhibition of breathing without complete arrest, bradycardia either of sinus origin or due to auriculoventricular dissociation, and increase in systolic and fall in diastolic pressure. These phenomena are in all respects comparable to those that result from the irritation of the nasal mucous membrane by water; they belong in the group of the inhibitory reflexes of the trigeminus and vagus. The reflex is regarded as explaining certain deaths of persons while swimming.

RUPTURE OF ALVEOLAR ECHINOCOCCUS CYST INTO THE PERITONEAL CAVITY AS A CAUSE OF SUDDEN DEATH. W. NEUGEBAUER, Centralbl. f. allg. Path. u. path. Anat. **58**:228, 1933.

A woman, 33 years of age, who had always been healthy, suddenly showed symptoms of peritonitis three days before her death. Abortion was suspected by the medical attendants, but at necropsy 1.5 liters of foul exudate was found in the abdomen. This material had apparently come from a ruptured cavity in the liver, the size of two fists. Rupture occurred into the omental bursa. The case is unusual in that no icterus was present before death. Usually compression of gall capillaries and widespread parenchymal necrosis lead to severe jaundice. There were no splenic enlargement and no enteritis. Echinococcus alveolaris ruptures much less frequently than does the hydatid form, and rupture of either is far more frequent into the subphrenic space or the pleural or pericardial cavity than into the omental bursa.

GEORGE RUKSTINAT.

MICROSCOPIC APPEARANCES OF THE GASTRO-INTESTINAL TRACT IN CHARRED BODIES. E. FRITZ, Deutsche Ztschr. f. d. ges. gerichtl. Med. **23**:19, 1934.

Microscopic examination of the gastric and intestinal mucous membrane showed perfect preservation of the surface epithelium with good nuclear staining. This state of preservation indicates that the heat had continued to act for a short time after death. In experiments on animals it was found that, when fixed by heat, mucous membranes resisted decomposition for three months after burial in earth.

FROM THE AUTHOR'S SUMMARY.

EXAMINATION OF THE WEAPON IN SUICIDE BY SHOOTING. BRUNING and WIETHOLD, Deutsche Ztschr. f. d. ges. gerichtl. Med. **23**:71, 1934.

In shooting with the muzzle against the skin fragments of blood and tissue nearly always enter the barrel, and consequently in doubtful cases examination of the weapon is of great importance.

SUICIDE BY CONSTANT CURRENT OF 220 VOLTS. W. MUNCH, Deutsche Ztschr. f. d. ges. gerichtl. Med. **23**:97, 1934.

Three cases of this rare form of suicide are described in detail, and twenty-nine reports in the literature of suicide by electric current are tabulated. In most cases the arrangement of the wires pointed directly to suicide.

DOUBLE MURDER BY THALLIUM. HERMAN KRSEK, Časop. lék. česk. **73**:40, 1934.

According to Krsek, this is the third report on homicidal thallium poisoning. The material used was Zeliopaste, which contains about 0.68 Gm. of thallium sulphate per tube. It is tasteless and is on unrestricted sale as a rat exterminator. The victims were the matrimonial partners of a middle-aged loving couple. The postmortem observations were practically negative in the man, who had received within a month an amount of thallium sulphate estimated to be between 0.68 and 2.05 Gm. The thallium content of his body was estimated at 1.33 Gm. The

woman's body contained about 1.62 Gm. It was exhumed eight months after death and was markedly better preserved than the average corpse in that cemetery. The lethal dose seems to be between 1 and 2 Gm. The changes which should arouse suspicion of thallium poisoning are clinical rather than pathologic. Thallium was mistaken for arsenic by the chemist in the case described by Kaps and Haberda. Gastro-intestinal symptoms accompanied by polyneuritis affecting especially the lower extremities (marked stabbing pains), hypo-acidity and trophic changes of the skin and especially loss of hair suggest this poisoning. In protracted cases—such as that of a woman who had received four doses in the course of three months—the physical symptoms may be dominated by mental disturbances of a hysterical or delirious character.

KAMIL SCHULHOF.

THE AIR CONTENT OF LUNG TISSUE. E. HUSTED and A. SAUGMAN, *Acta path. et microbiol. Scandinav.* **11**:227, 1934.

The authors have devised a modified floating test whereby the lung tissue is placed in water in a sealed container connected with a manometer and by a three-way stopcock with apparatus for increasing and decreasing atmospheric pressure. By means of a mathematical formula and a table it is possible to determine the percentage of air in a given piece of lung tissue. The authors tested fifty-three specimens of lung tissue from twenty-three persons, including fifteen newly born. Normal fully expanded lung could not be made to sink in the water. The air of the lungs has to be reduced considerably before an overpressure of from 500 to 600 mm. is able to sink the tissue. A partly atelectatic piece of lung from a newborn infant who died fourteen hours post partum sank in the ordinary floating test, but could be made to rise with a pressure of —130 mm. By this simple test it is possible to demonstrate the presence of air in lung tissues when the ordinary floating test shows apparently air-free lungs.

JACOB KLEIN.

EXPERIENCES WITH FATAL HEAD INJURIES. F. HARBITZ, *Norsk mag. f. lægevidensk.* **95**:353, 1934.

Harbitz studied 115 fatal head injuries of which 50 were homicidal, 40 accidental and frequently associated with drunkenness and 25 due to automobile injuries. He has a detailed tabulation of 20 cases in which in addition to skull fracture there were hemorrhage between the brain coverings, tearing of the brain cortex and hemorrhage of the brain in the central ganglions, pons and medulla oblongata. These injuries were especially common after automobile accidents, and most marked in the temporal region. In 83 bodies there were: large epidural hematomas in 13, either with or without a skull fracture; extensive subdural hemorrhages in 19; large hemorrhages in the arachnoid in 14; tears and hemorrhages of the brain cortex, and in 12, small hemorrhages in the interior of the brain, mostly in the line of direction of the acting force. Harbitz reports five deaths from commotio cerebri. He found no constant anatomic alterations in this condition. He is of the opinion that there is no sharp boundary between commotio cerebri and contusio cerebri and does not believe the cause of the former or the reason for its causing death is established. He has not seen hemorrhage in the interior of the brain in the central ganglions or pons or medulla, with breaking into the third or fourth ventricles due to violence to the head, in the absence of skull fracture, hemorrhage into the brain coverings or tearing of the brain cortex with some small outer damage. Three cases of combined traumatic and spontaneous bleeding in the brain were encountered in men from 53 to 61 years old. The hemorrhages regarded as spontaneous were situated in the pons. He warns against drawing conclusions in such cases because emphasis on the traumatic features may have widespread medicolegal implications. Two deaths in men known to have hypertension, but who had injuries to the face as well as intracerebral hemorrhages, were regarded as due to natural causes. It was assumed they had the intracerebral lesion first and then fell, injuring the face.

GEORGE RUKSTINAT.

# Society Transactions

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## BUFFALO PATHOLOGICAL SOCIETY

*Regular Meeting, May 12, 1934*

KORNEL TERPLAN, *President, in the Chair*

ACUTE LEUKEMIC RETICULO-ENDOTHELIOSIS. JOHN LOESCH.

Five weeks prior to his admission to the hospital a white man, aged 30 years, an iron-worker in an automobile factory, noted the development of hemorrhagic areas in the skin over the entire body. Three days later his throat became severely sore and his neck swollen. Following the appearance of these conditions, which grew progressively worse, jaundice was noticed; bleeding from the mouth and rectum occurred. Exposure for thirty-six hours on an ice flow in Lake Erie antedated the patient's illness.

On physical examination the skin, in addition to the hemorrhagic lesions, showed, especially on the chest and abdomen, circular elevated nodules of sizes up to that of a dime. The liver projected four fingerbreadths below the costal margin. The spleen reached into the pelvis. The inguinal nodes were enlarged. The laboratory work showed: erythrocytes, 2,580,000, and leukocytes, 12,200, per cubic millimeter, with adult polymorphonuclears 3 per cent, lymphocytes 27 per cent and monocytes 69 per cent. In the last group, the nuclei were round or elliptic in 28 per cent, slightly indented in 23 per cent, deeply indented in 10 per cent and lobulated in 8 per cent. Mitotic figures were noted. Frequently the cytoplasm of the monocytes contained fragments of red blood cells. The blood platelets appeared diminished in number. The bleeding time was fifteen minutes; the clotting time, twenty minutes. A blood culture was positive for hemolytic staphylococcus. The clinical diagnosis was: acute leukemia and septicemia with multiple abscesses.

The chief postmortem observations comprised: marked jaundice; multiple hemorrhages and circular elevated nodules in the skin; petechial hemorrhages of the uterus and serous membranes; marked enlargement of the liver; marked enlargement of the spleen; enlargement of all the lymph nodes, especially those in the left tracheobronchial and inguinal groups; numerous white areas, 0.5 cm. in diameter, scattered throughout the myocardium, papillary muscles and kidneys; 250 cc. of slightly hemorrhagic fluid in the peritoneal cavity, and yellowish-red bone marrow.

Histologic examination of the liver, spleen, various lymph nodes, skin, bone marrow, kidney, myocardium and tonsil, showed a uniform proliferation of large mononuclear cells with abundant cytoplasm and very often with distinctly indented nuclei. The picture, especially in the myocardium, resembled closely a blastomatous infiltration. Pathologically, this condition, the intravital blood picture of which corresponded to a so-called monocytic leukemia, was diagnosed as acute leukemic reticulo-endotheliosis. The history of exposure was considered in a discussion of the etiology.

### DISCUSSION

K. TERPLAN: This case evoked considerable discussion in the matter of differential diagnosis, but from a histologic analysis of Dr. Loesch's slides I feel that an acute myeloblastic leukemia can be ruled out. The nuclei of the cells shown possess a structure that is generally recognized as belonging to those of reticulum cells or monocytes. I am especially impressed by the blastoma-like infiltration of the myocardium. Dr. Loesch's case is certainly different from the instances

reported in the literature as acute reticulo-endotheliosis, in which no increase of monocytes was noted in the circulating blood. In these cases the histologic picture also resembled that of a reactive hyperplasia of reticulum and endothelial cells. In some, the unusual reaction was explained on the basis of an infection.

ACUTE LEUKEMIA FOLLOWING ADMINISTRATION OF ARSPHENAMINE. S. L. VAUGHAN, K. TERPLAN AND S. SANES.

Cases of acute apparently lymphatic leukemia following arsphenamine therapy are relatively rare.

A white man, aged 54 years, was admitted to the Buffalo General Hospital, complaining of a swelling on the left side of the face of five days' duration. He had finished an intensive course of sulpharsphenamine injections for tertiary syphilis three months previously. Physical examination revealed swelling of the parotid gland resembling acute parotitis, primary optic atrophy and palpable inguinal lymph nodes. The liver and spleen were not felt. The temperature was 100.6 F. The urine contained 0.01 mg. of arsenic per hundred cubic centimeters.

The blood revealed 2,200,000 erythrocytes per cubic millimeter, of uniformly large size, predominately oval shape and even staining. Reticulocytes were 0.2 per cent. The hemoglobin content was 56 per cent. The leukocyte count was 2,200 with 2 per cent filament neutrophils, 1 per cent eosinophils, 84 per cent lymphocytes and 13 per cent monocytes. The thrombocyte count was 123,000, the Wassermann reaction 4 plus.

On intramuscular injection of liver extract there were a rise in the total leukocyte count to 4,700 and a fall in the thrombocyte count to 86,000, but no significant changes in the erythrocyte or differential counts.

Five daily injections of pentose nucleotide were followed by further rise in the leukocyte count to 5,900, due to the appearance of cells regarded as young monocytes in the concentration of 19 per cent. The peroxidase reaction on these cells was negative. The erythrocytes showed polychromatophilia and poikilocytosis for the first time.

After an afebrile period of two weeks the temperature began to rise again and remained elevated thereafter. Exudate which was obtained from a few pustules that had developed on arms and face was quite fluid, and contained very few polymorphonuclear cells. The predominating cells closely resembled those seen in the blood, although many of them gave a strongly positive peroxidase reaction. *Staphylococcus aureus* was recovered in pure culture.

During the next month the leukocyte count rose to 55,000, and after a temporary fall, to 124,000. Polymorphonuclear cells remained less than 1 per cent, lymphocytes 30 per cent, mature monocytes 9 per cent, and the cells described previously 60 per cent. In the terminal stage the last cells became less granular, but a small percentage of them gave a positive peroxidase reaction. The anemia progressed slightly, and a thrombocytopenia of 25,000 developed. An ulcer appeared on the gum. Lymph nodes became palpable for the first time in the neck and axillae. Death occurred ninety-four days after admission.

Autopsy showed: marked generalized anemia; distinct hemosiderosis of the liver; marked "tigering" of the heart, with dilatation; minute whitish infiltrates scattered regularly throughout the cut surface of the liver, which was not conspicuously enlarged; enlargement of the spleen (weight, 280 Gm.), with entirely obscured structures; anemic jelly-like appearing bone marrow in the spine and the sternum; fatty marrow in the femur; moderate turgescence of the penis suggesting priapism; moderate enlargement of the peripancreatic, periaortic, pelvic and inguinal lymph nodes; distinct yellowish-white clots in the large veins and heart; rather watery blood issuing from cut vessels.

Histologically, the spleen, liver, lymph nodes, kidneys, corpora cavernosa, heart muscle and bone marrow exhibited a uniform proliferation chiefly of immature large round cells which were considered as lymphoid elements including lymphoblasts. The distribution of the infiltration in the liver and kidney pointed to lymphatic leukemia. Practically not one of the cells showed granular cytoplasm.

There was no proliferation of reticulum cells. No recognizable myeloid elements were noted.

Whether the true leukemic stage is a compensatory effort to a marked suppression of bone marrow, or whether it represents a direct response to the toxic agent, sulpharsphenamine, can hardly be decided in the case here reported.

**PRIMARY CARCINOMA OF THE STOMACH—GROSSLY UNRECOGNIZABLE—WITH EXTENSIVE METASTASES TO THE BONE MARROW PRODUCING MARKED INTRAVITAL ERYTHROBLASTOSIS.** K. TERPLAN AND S. L. VAUGHAN.

This case is presented because the results of clinical and hematologic examinations were rather suggestive of a malignant process of the gastro-intestinal tract with metastases to various parts of the bone marrow. Yet gross inspection at autopsy failed to disclose clearly the site of the primary tumor.

A Polish woman, 42 years of age, was admitted to the General Hospital, Jan. 31, 1934, complaining of weakness and loss of 62 pounds (28.1 Kg.) in weight for one year. For three months prior to her entry the patient suffered with anorexia and generalized body pains.

Physical examination revealed an emaciated cachectic patient, with marked general pallor and with an indistinct yellowish cast to the skin and conjunctiva. Lymph nodes not exceeding the size of a pea were palpated in the neck, axillae and groin. The heart was enlarged to the left; a systolic murmur was heard over the apex and the base. The liver extended from the seventh interspace to the level of the umbilicus in the anterior axillary line. It was tender. The spleen reached to the level of the umbilicus. A moderate amount of fluid was in the peritoneal cavity. A number of lentil-sized discrete nodules were felt on the right pelvic wall by rectal examination.

A colonic injection showed no evidence of neoplasm. The urine contained abnormally large amounts of urobilinogen. The gastric contents were negative for hydrochloric acid after injection of histamine. Evidences of pyloric obstruction were not found. The feces were negative for occult blood on two occasions; the color was dark brown. The blood serum contained 1.4 van den Bergh units of bilirubin. The red blood count was 1,700,000; the hemoglobin content was 44 per cent. The cells varied considerably in size with the average distinctly greater than normal. Poikilocytosis and polychromatophilia were marked. Staining was normally intense; stippling was present. Erythroblasts numbered 6,200 per cubic millimeter. Of the erythrocytes, 17 per cent were reticulated. The leukocytes numbered 8,700 per cubic millimeter, with myelocytes 18 per cent, juvenile forms 14 per cent, band forms 27 per cent, filament forms 15 per cent, eosinophils 1 per cent, lymphocytes 16 per cent; lymphoblasts 2 per cent, monocytes 6 per cent and Türck's cells 1 per cent. The thrombocytes, many of which were dime-sized, numbered 30,000. The clinical picture suggested malignant tumor, probably in the gastro-intestinal tract. The profound anemia with thrombocytopenia accompanied by a tremendous regenerative effort in both the erythrocytic and white cell series was thought to be diagnostic of sudden extensive encroachment on active bone marrow tissue, such as one might expect from tumor metastases.

At autopsy there were marked generalized marasmus and anemia. The stomach appeared distinctly contracted. It showed slightly increased mucoid secretion and rather prominent folds. The mucosa was entirely intact. Grossly, infiltration at any part of the wall of the stomach could not be noticed. Only at the upper part of the lesser curvature were the serosa and mesogastrum firm and thickened. There were also a few small slightly cystic nodules in the omentum and rather many minute metastases in the lungs and bronchomediastinal lymph nodes. Both ovaries were moderately enlarged. The vertebral bodies, especially those of the lumbar spine, were diffusely invaded by soft metastatic tumor. The histologic type was adenocarcinoma, with marked mucoid production. Sections taken from different parts of the stomach revealed that the primary tumor originated apparently in the upper part of the fundus near the lesser curvature. The superficial layer of mucosa was intact. There was a slight disorder of the

glands in the depth of the mucosa with distinct infiltration of the lymph spaces. The submucous spaces were entirely filled with carcinomatous cells. Here and there the carcinomatous cell cords extended through the muscularis mucosae. Diffuse invasion also was seen in the muscle layer and subserous spaces, even in areas more distant from the lesser curvature. Metastases from the omentum, lung and bronchomedastinal lymph nodes showed the same histologic type throughout; the ovaries showed the typical picture of so-called Krukenberg tumor. The liver and spleen exhibited unusually marked myeloid metaplasia; the histologic observations were hardly different from those seen in true myelogenous leukemia.

This is the second case in which, so far as we know, primary carcinoma of the stomach produced no structural alteration of the entire stomach which could be grossly detected. The first case was reported several years ago from Prague by one of us (Terplan, K.: *Bcitr. path. Anat. u. z. allg. Path.* 87:229, 1931).

#### STUDIES OF BLOOD CALCIUM, POTASSIUM AND CHOLESTEROL IN DOGS FOLLOWING KIDNEY DAMAGE, EXPERIMENTALLY PRODUCED. JOHN LOESCH.

*Series A.*—In dogs on a standard diet the kidney was damaged by ischemia. This was accomplished by clamping the renal pedicle for a certain duration at certain intervals. The calcium figures were normal except in the terminal stage, when the changes were not consistent. In some animals the values decreased, occasionally to 50 per cent of the initial ones. In other animals the figures were increased. The potassium, however, was uniformly elevated and reached highest values in the terminal stage. The potassium-calcium quotient rose simultaneously with the foregoing changes.

*Series B.*—In dogs on a standard diet as much as three fourths of the total renal parenchyma was removed. No significant changes occurred in potassium, calcium or the potassium-calcium quotient. When the protein in the diet was increased, the potassium always tended to rise, and the highest figures were found in the uremic stage. As the calcium remained normal or decreased slightly, the potassium-calcium quotient increased.

In an attempt to establish a relationship in each series between calcium, potassium and the potassium-calcium quotient on the one hand and the nonprotein nitrogen, plasma protein content and plasma percentage on the other hand, it was found that of the first three the potassium and potassium-calcium quotient alone varied in direct proportion to the last three.

As the cholesterol figures showed great variation in both series definite conclusions could not be drawn from them in any of the experiments.

The foregoing work was carried out simultaneously with the studies of plasma proteins reported before the Buffalo Pathological Society, at the meeting of Nov. 14, 1932 (*Arch. Path.* 14:901, 1932).

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#### NEW YORK PATHOLOGICAL SOCIETY

*Regular Meeting, May 24, 1934*

WILLIAM C. VON GLAHN, *President, in the Chair*

MITRAL STENOSIS WITH INTERAURICULAR INSUFFICIENCY. SEATON SAILER (by invitation).

A case of mitral stenosis with interauricular insufficiency is reported in a white man of 67 years. This brings the total number of cases reported to twenty-seven. It is the third reported case occurring in a male. A clinical diagnosis of this combination of lesions is possible, being determined mainly by roentgen and fluoroscopic examination. The criteria are as follows: There is an enlarged,

transverse and sabot-shaped heart. The abnormality is caused by a great enlargement of the right auricle and ventricle with rotation of the left ventricle posteriorly, the right ventricle forming almost the entire anterior aspect of the heart, including its apex. The pulmonary artery is greatly enlarged throughout, and its hilar branches form discrete rounded shadows. The aorta is hypoplastic. Fluoroscopic examination shows an upward and outward counter-clockwise movement of the right ventricle during systole with uncovering of its inferior border, normally hidden in the diaphragm. The hilar shadows of the pulmonary artery show an expansile pulsation. Auscultatory signs of mitral stenosis or of a patent interauricular septum are corroborative, but not constant. The electrocardiogram shows right ventricle preponderance and notched R waves in all leads. The P and T waves show no distinct changes.

In the case which I shall now report the heart weighed 700 Gm. A moderate degree of stenosis of the mitral valve was present. It measured 9 cm. in circumference. The right ventricle measured 1.5 to 1.7 cm. in thickness. The pulmonary artery was symmetrically enlarged, measuring 11.5 cm. in circumference to its point of bifurcation. Its ramifications throughout the lung were greatly enlarged. The aorta measured 6.5 cm. in circumference. Microscopically both vessels showed slight degenerative changes of the medial muscle cells and fraying of the elastic tissue. A defect 3 cm. in diameter was present at the lower border of the auricular septum above the septal flap of the tricuspid valve, the endocardial lining of this valve being continuous through the foramen with that of the posterior mitral curtain. The right auricle was markedly dilated and hypertrophied; the left, moderately involved. A slight fusion of the commissures of the aortic valve was present, with resulting hypertrophy of the wall of the left ventricle. Death was caused by confluent bronchopneumonia involving the middle and lower lobes of the right lung and the lower lobe of the left lung.

#### SYNOVIOMA: A REPORT OF THREE CASES. LEILA CHARLTON KNOX.

Under the name "synovioma" may be designated a special group of malignant connective tissue tumors occurring in the extremities and arising from the synovia of joints, tendon sheaths and bursae. These may be either intra-articular or extra-articular. The intra-articular group are apparently more slowly growing and inclined to be nodular for a longer period of time, but like those external to the joints, they recur and frequently metastasize widely. These growths have been most extensively described by the French school.

Three cases are presented. The first occurred in a young woman, the tumor arising from either a bursa or a tendon sheath on the ulnar aspect of the right elbow. It caused pain for one and a half years, and then grew fairly rapidly for another year and a half. Amputation was done in August 1921. The patient remained in good health for over six years, but died with extensive recurrences in the shoulder, chest and cervical nodes in 1928.

The second patient was a man 33 years of age, first seen in June 1929, with a tumor in the right popliteal space, which had originated apparently around the tendon sheaths and had grown slowly for six months until it was removed locally for microscopic examination. A midthigh amputation was done a few days later, but the patient died early in 1932, with roentgen and clinical evidence of metastatic tumor of the right lung.

The third patient is a man 24 years of age who had a tumor on the plantar surface of his left foot which had been causing pain and disability for two and a half years. The growth was found under the plantar fascia and surrounding the tendons. It was infiltrating diffusely, and was operated on. The first diagnosis was hemangioma. A diagnosis of malignant tumor was made microscopically, and the leg was amputated three weeks later. The patient is now, two years after the operation, without clinical evidence of metastasis.

The microscopic appearance of these tumors is that of a small spindle-cell sarcoma with a tendency toward mucinous degeneration of the cytoplasm. The

nuclei are not extremely hyperchromatic, but the number of nuclei in any field is large. A characteristic feature not present in every section, but probably present in every tumor, is the organoid structures superficially resembling epithelium in which the altered fibroblasts tend to surround a lumen, although no basement membrane, epithelial secretions or cilia are found. These structures probably represent the tendency of the tumor to reproduce in some of its cells the alterations that take place when a synovial membrane forms about a free cavity.

Except for the presence of these pseudoglandular structures, the tumors closely resemble some types of periosteal sarcoma, from which they should be differentiated. Roentgenograms of the bones show no involvement in the early stages, and only pressure atrophy and absorption or secondary invasion in the late stages.

They should also be differentiated from xanthomas of the tendon sheaths, frequently designated as giant-cell sarcoma, the xanthomas being benign growths, never malignant. The effects of radiation on the tumors have not been established, as so few of them have been recognized and treated with measured quantities for any adequate period. Amputation is frequently necessary, but undoubtedly some patients have been cured with less radical treatment.

#### DISCUSSION

ALFRED PLAUT: How do such tumors react to radiation? I should like to ask also about the clear substance of some of the cells: Does it give any metachromatic reaction with mucicarmine or thionine?

FRANCIS CARTER WOOD: I have been following for a number of years a patient with a tumor of the popliteal space probably belonging in this group though it was purely spindle cell in type. The sections were not very good, and I have doubted the clinical diagnosis somewhat because the patient is still alive. But it has been only five years since his operation, so that one still has a chance to prove or disprove the presence of malignancy. There was none of the alveolar structures that Dr. Knox's tumors showed. The growth was very radically removed and then the involved region was heavily irradiated, with rather disastrous results, for when I saw the patient, he was suffering more from his irradiation than from his tumor. He had a large ulcer over the popliteal space where the radiation had been applied, and there was some question of further surgical intervention, which I opposed. I saw him a few weeks ago, and he was in perfect health; the ulcer had finally healed. His was a very puzzling case; the slides were shown to a number of pathologists, and a great variety of diagnoses were made.

It is important that Dr. Knox has given a picture of this condition so that cases can be recognized promptly. I should not think the growths would be radiosensitive, any more than the other spindle-cell sarcomas are. One has to differentiate tumors of this type clinically very carefully from the other group of tendon sheath tumors, that is, the giant-cell tumors, which are merely inflammatory reaction products; in the older literature the latter type is often confused with tumors of the type just described, so that the older pathologists thought they might be melanoma. That group of tendon sheath tumors never evinces malignancy, though they may recur locally.

LEILA CHARLTON KNOX: Very little has been learned from special stains. There is no mucoid secretion, nor any metachromic reaction with the mucicarmine stain. The eosin and hematoxylin stains show better than others the relation of the cells to one another.

With regard to radiosensitivity there are very few observations, because the total number of treated persons is so few, and the technic of radiotherapy has been altered so many times in the last ten years that even the few observations which have been made may not be significant for the group. Smith (*Am. J. Path.* 3:355, 1927) cites two cases in which such treatment was given, and the cutaneous and pulmonary metastases were very considerably improved by the x-rays. That was only a few years ago. One of Dr. Wagner's patients, the one with the large intra-articular tumor, was treated by Dr. Wood for some time before he

consented to have his leg amputated, and during that time the tumor was slowly growing into the head of the tibia. Therefore that particular tumor was not radiosensitive. In the second case that I cited the metastases in the chest were treated with radiotherapy. The patient showed no improvement. That may have been because the tumor was rather extensive at the time treatment was begun, but tumors of this type are certainly not highly sensitive to radiation.

ALFRED PLAUT: I suppose it is possible by x-ray picture to make a correct differential diagnosis between a bone sarcoma on which one would not do a biopsy, and a tumor such as this, in which one would like to have a biopsy first, in order to decide on amputation.

LEILA C. KNOX: The question probably has to be settled by biopsy. There was some erosion of both condyles of the femur in one patient and of the heads of the humerus and radius in another of these patients, but it seemed to be purely secondary. The tumor was invading the bone as it invaded the soft tissues, and the outer part of the cortex was gone. But the shaft of the bone was still there. The large intra-articular tumor usually makes a cavity for itself. In many of the cases the growth has clinically resembled tuberculosis of the joint. The older French and German literature discloses that many of the patients were believed to have tuberculosis of the joint because of the spindle-shaped swelling and the semi-fluctuation.

HIRSUTISM, HYPERTENSION AND OBESITY ASSOCIATED WITH CARCINOMA OF THE SUPRARENAL CORTEX AND AN INDETERMINATE PITUITARY ADENOMA.  
IRVING GRAEF and (by invitation) ANTONIO ROTTINO and JOSEPH J. BUNIM.

The case of a patient exhibiting the symptoms of hypertrichosis, rapidly acquired obesity and hypertension is presented: H. B., an unmarried white woman of 19 years, was studied on the Third (New York University) Medical Service of Bellevue Hospital from Nov. 13, 1933, until death, Jan. 31, 1934. The complaints were of progressive weakness and edema of the feet of nine months' duration.

The onset of menstruation was at 12; the flow recurred every thirty days and was painless and moderate, lasting seven days. There were no menses during March or April of 1933 or during the last two months of her life.

The hypertrichosis was sufficiently marked to require shaving of the face in the last four months.

In November 1933 the blood pressure was 124 systolic and 90 diastolic; subsequently the systolic pressure ranged from 168 to 180, and the diastolic, from 125 to 138.

The patient was of hypersthenic habitus, weighed 156½ pounds (71 Kg.) and measured 65 inches (165 cm.). The body had a peculiar pungent, sweaty odor.

Long, broad striae were seen over the abdomen and upper part of the thighs; smaller ones over both mammae and the upper part of the arms.

Hypertrichosis consisting of tawny yellow hair was noted over the face, upper lip and both extremities. There was female distribution of the pubic hair. The clitoris was of normal size. There was marked edema of both legs.

No voice changes were noted. The Aschheim-Zondek test was negative. The basal metabolic rate was -7 and +3 per cent. The Wassermann test was negative. Urinalyses showed proteinuria (from + to ++++). The specific gravity ranged from 1.005 to 1.015. No reducing or acetone bodies were detected. Dextrose tolerance was diminished. Roentgen study of the chest showed several circumscribed areas of increased density in both pulmonary fields, suggesting metastatic new growths.

Dr. Robert T. Frank of Mount Sinai Hospital made hormonal studies of the patient's blood and urine. He reported: "The urine contained 5,000 mouse units of female sex hormone per liter; as a control, the Friedman test was done with the same urine on two rabbits, with negative results. There was no excess of anterior pituitary-like factor in the urine. The venous blood contained 1 mouse unit in 40 cc., which is a quite normal finding."

Six days before death, the temperature rose to 103 F., and a large abscess was noted in the upper part of the left thigh, from which hemolytic streptococci were isolated. The striae of the lower part of the abdomen began to suppurate, and the patient died in coma.

Autopsy was performed two hours after death. The external examination conformed to the description given. The essential pathologic observations follow: The right kidney was found displaced downward by a large tumor situated at its upper pole. Posteriorly the tumor completely compressed the inferior vena cava and was firmly attached to the right lobe of the liver. Section showed an irregular lobulated appearance. Some areas were butter-yellow; some, the seat of recent and old hemorrhage. There was deep invasion of the right lobe of the liver. The inferior vena cava from its opening in the right auricle was filled with soft tumor deposits.

In the lungs were numerous metastatic nodules. The heart was not enlarged; the wall of the left ventricle was of normal thickness. The left suprarenal gland was small and wrinkled. Both ovaries were small and firm. Other organs did not show gross changes. On dissection of the pituitary body no gross changes were found. It measured 13 mm. in the anteroposterior lateral, 14 mm. in the lateral, and 6 mm. in the vertical, plane.

Microscopic sections of the tumor revealed a new growth consistent with the observations in carcinoma arising in the cortex of the suprarenal gland. Tissue fixed in absolute alcohol and stained by Best's method showed no glycogen. The thyroid gland appeared to be in a resting stage. Both ovaries were relatively bloodless and showed an unusual amount of progressive atresia of the follicles in all stages. Sections of the endometrium showed atrophy of the glands and marked edema of the outer two thirds of the mucosa. In the left suprarenal gland the medulla appeared scanty, the cortex narrow and the zona reticularis atrophic. The pineal body showed no changes. The other organs with the exception of the pituitary gland were not significantly altered. After fixation in Zenker's fluid the halves of the pituitary body were embedded in paraffin.

Serial sections of the pituitary body, 5 microns thick, were prepared. In one block a circumscribed miliary nodule was found, which was oval-shaped and measured 1 mm. by 850 microns. At first, from hematoxylin and eosin sections alone it was thought that this was a basophilic adenoma, but special cytoplasmic stains, such as Sevringshaus' method, Bailey's method employing acid violet and Altmann's aniline fuchsin, Mallory's aniline blue, the Weigert method employing gentian violet, iron hematoxylin, Heidenhain's azo-carmine, and a new method introduced by Soas and Csizek, revealed features as follows: There were a few eosinophils and an occasional intact basophil in the periphery of the nodule, but the vast majority of the cells showed an ill-defined cytoplasm in which no chromophilic granules were demonstrable. While the majority of the cells tended to take a feeble purplish tone with hematoxylin, ordinary immersion in 1 per cent aqueous eosin for one minute gave them a reddish tint. Reticulum stains (the method of Foot and Foot) revealed the absence of the normal reticular stroma within the nodule and a poorly defined capsule which demarcated the nodule clearly from the adjacent tissue. The configuration of the adjacent tissue showed compression of the cords of the nodule.

The nature of this nodule was difficult to determine; it partially satisfied the criteria given by Kraus in his study of anterior pituitary adenomas. It possessed a capsule and compressed the adjacent tissue, and for the most part it was composed of chromophobic cells, degenerating and poorly defined. The state of these cells was unusual. Many multinucleated forms were seen, and while the eosinophils were well preserved, the cells here as well as the occasional basophils at the periphery showed profound cytoplasmic and nuclear changes.

These changes were not confined to the nodule, however, and were seen in the basophils elsewhere in the pars distalis. These consisted of marked nuclear swelling, dissolution, vesiculation, lobulation, amitotic division, extrusion of nucleoli, wrinkling of the nuclear membrane and the cytoplasmic membrane, and loss of

cytoplasmic granules. The eosinophils remained well preserved. The sections of the pars nervosa showed no invasion of epithelial elements, and the pars tuberalis also seemed normal.

Interpretation of these observations in the light of present knowledge is difficult. Pituitary disorders are known to influence the suprarenal cortex; for example, aplasia of the cortex in anencephaly, P. E. Smith's experimental atrophy of the suprarenal cortex in hypophysectomized rats and cortical adenomatosis of the suprarenal gland in Putnam's dog rendered acromegalic by injections of anterior pituitary extract. There is clinical evidence of the suprarenal-pituitary relationship in acromegaly and in infantilism. Of great interest are the eighteen cases of pituitary basophilism (Cushing) now recorded. Nine of the patients showed cortical hyperplasia or hypertrophy of the suprarenal gland and one an adenoma of the suprarenal cortex.

While the pituitary adenoma in the present case may have been a coincidental finding, to decide this even tentatively is difficult. It is pointed out that there are no satisfactory statistical data on the incidence of pituitary adenomas in the general population independent of pituitary disorders. A synthetic series based on Erdheim's, Kraus' and Kiyono's serially sectioned pituitary glands shows that adenomas are rare in youth. Evidence to show more than a coincidental relationship between the suprarenal and the pituitary gland in this case is submitted from the literature: 1. A case reported by Oppenheimer and Fishberg (*Arch. Int. Med.* **34**:631, 1924) of a man of 24 with a suprarenal cortical tumor and hypertension. The authors noted that their patient had an acromegalic appearance. 2. A case reported by Long and Gray in the *Medical Journal and Record* (**119**:38, 1924) of a metastasizing suprarenal cortical carcinoma in a man, aged 45, in whom an acromegalic appearance developed while he was under observation. In the first case no statement was made concerning histologic examination of the pituitary gland. In Long and Gray's case, casual sections were reported to show an overgrowth of eosinophilic and chromophobic cells, also epithelial cell invasion of the pars nervosa. 3. A case reported by Mathias (*Virchow's Arch. f. path. Anat.* **236**:446, 1922) of an 18 year old girl with hypertrichosis since the age of 3 who died with a metastasizing suprarenal cortical neoplasm. Serial sections revealed a marked increase of eosinophilic elements and, further, an overgrowth of the chief cells which in one place had the character of an adenoma.

If, as Cushing has shown, tumors of the pituitary gland are frequently associated with hyperplasia of the suprarenal cortex, it is suggested that the evidence presented from the literature, as well as in our own case, tends to show that tumors of the suprarenal cortex may be associated with changes or manifestations of changes in the pituitary body.

#### DISCUSSION

ROBERT T. FRANK: I am thankful for the chance to see this patient during life, and later, after death, to obtain enough of the organs to make extracts, because I had a similar case which I studied for over a year and a half. Without taking any position in the discussion of whether this is of primary basophilic or of suprarenal origin, I may say that the syndrome is very striking. The case I had will be reported in detail by Dr. Oppenheimer and myself; the patient died on Dr. Oppenheimer's service, but I am particularly interested in showing some of the biologic changes which have taken place.

The patient had the typical appearances of what Cushing described as basophilic adenoma: hirsutism, particularly noticeable on the chest and face; a pendulous abdomen, and small legs and arms in contrast to the thick, fleshy abdomen. The striae were very marked, but not so marked as in the case presented by Dr. Graef. I was so convinced that the patient had a basophilic adenoma that I took occasion at the meeting of the Congress of Surgeons and Physicians in 1933, at which Dr. Cushing was presiding, to present the case as one of basophilic adenoma, and as the first in which the hormones had been studied. This slide shows the female sex hormone in the blood. It is that of the normal female. It also shows the prepituitary secretion in the blood, which likewise shows no abnormality, but the

excretion of the estrogenous substance in the urine was tremendous. This small area shows the amount of female sex hormone which is excreted by a normally fertile menstruating woman during a cycle, while this patient excreted as much as 17,500 mouse units of sex hormone in the course of three days. The ordinary female excretion is about 1,500 mouse units in thirty days. The tremendous excretion led us to make numerous tests for pregnancy, the results of which proved to be negative.

Dr. Bunim asked me how a patient who has atrophic ovaries, as our patient and Dr. Graef's had, with an atrophic endometrium, can produce such a tremendous amount of female sex hormone that it is split over through the kidneys and exceeds that found in pregnancy, and I cannot answer that question. The nearest approach to an answer that I can give is a purely hypothetic one, and it is permitted to me through the study of the extracts. The tumor itself on extraction gave twice as much female sex hormone to the gram as indifferent tissues such as the spleen, of which I likewise had material. The difference is not tremendously striking. Implantation gave no definite results. This is the hypothesis: The suprarenal gland can produce estrogenic substances under these conditions. The mere fact that this large tumor was comparatively poor in estrogenic substance, though much richer than the other organs, does not necessarily mean that this hypothesis is wrong, because if one takes a portion of a corpus luteum one gets very little female sex hormone there, although one knows that it excretes it in large amount; in other words, it does not correspond to the thyroid gland, which is a storage organ, but to an organ which is in very close relation to the capillaries, and there is an immediate distribution of its product. I put this as an open question, because I have no proof of it. Do the carcinomas and the adenomas of the suprarenal glands differ? Through the kindness of Dr. Wilder and Dr. Snell of the Mayo Clinic I was able to obtain some urine from two persons with adenoma, non-malignant, who were successfully operated on a year ago at that clinic. All their symptoms have receded since operation, and the results of the tests for hormone were negative, so that the question remains unsettled.

From a diagnostic standpoint this overexcretion may be of extreme importance. X-ray pictures of the sella turcica yield no evidence. The pituitary adenomas are so small that they do not change the bone. On the other hand, if one finds overexcretion in the urine, that may be a test, and I have offered it as a test. An article on the subject has been published in the *Proceedings of the Society of Experimental Biology and Medicine* (31:1014, 1934), because such cases are so rare that the profession at large must try it out and see. For example, a thorough study of the urinary tract in our patient showed no displacement of the kidney as in Dr. Graef's patient, although a large carcinoma was found. I have had the opportunity of examining the urine in several cases which resembled cases of the basophilic type, and it has been normal in all of them. I hope, as the test becomes known, that it will be tried by others, and that it will help in differentiation.

ALFRED PLAUT: Dr. Graef gave me an opportunity to study the slides of the hypophysis in his case. About the suprarenal tumor I have nothing to say. Dr. Graef has stressed the fact that little is known about the influence of the suprarenal gland on the hypophysis. In this connection I should like to draw attention to the absence of important hypophyseal changes in Addison's disease. This at least shows that a complete or nearly complete destruction of the suprarenal gland does not necessarily have a morphologic manifestation in the hypophysis.

So far as the hypophysis in Dr. Graef's case is concerned, I should like to divide the question into two parts: first, that of the changes in the cells and the nuclei, second, that of the adenoma. I agree with Dr. Graef that one cannot make any definite statement about the cellular changes. While Dr. Graef was demonstrating his slides it occurred to me that very similar changes had been seen in the hypophysis of a patient with severe endocrine disturbances. This patient also had a large adenoma of the hypophysis. The patient was a pregnant woman, 25 years of age, who died of cerebral hemorrhage during labor. She was very obese; she had had late menarche; she had splanchnomegaly and some changes similar to von Recklinghausen's neurofibromatosis.

In my opinion the adenoma in the anterior lobe of the hypophysis of Dr. Graef's patient is of no importance. Dr. Graef has said that adenomas of the anterior lobe are very frequent. In the hospital with which I am affiliated we went over the last one hundred and ten autopsies in which the hypophysis was removed and found five definite large adenomas. Only four of the one hundred and ten hypophyses were examined in serial sections. In seventy-nine cases only a dozen slides or less were examined. From each of twenty-three hypophyses between twelve and fifty slides were available; in only four cases between fifty and one hundred slides were available. Only one of the five adenomas was found during serial examination. Three of the others were found in hypophyses from each of which little more than a dozen slides were examined. The fifth was detected while fifty slides of the hypophysis were being examined. Our percentage is still much lower than that of Erdheim and of E. J. Kraus. They have found adenomas in 10 per cent of their hypophyses.

Out of our five cases of adenoma only the one that I have mentioned concerned a patient with endocrine disturbance. The other adenomas were found accidentally, one in a woman 67 years old with carcinoma of the kidney, one in a middle-aged man with chronic lymphatic leukemia and lobar pneumonia, one in an old man with coronary sclerosis, and one in a woman 40 years old with metastases from carcinoma of the breast and multiple abscesses of the kidneys.

None of the five adenomas was basophilic. One is easily led into making a diagnosis of basophilic adenoma from a casual examination of the section stained with hematoxylin and eosin. The number of nuclei is often large on account of the small size of the cells; thus the whole adenoma makes a large blue dot in the slide and misleads one into a diagnosis of basophilic adenoma. (Lantern slides were demonstrated.) The adenomatous tissue and the normal tissue of the anterior lobe are continuous in most cases, just as one sees tumor tissue and normal tissue merging into each other in myoma of the uterus, for instance, or in goiter.

In the woman who died of metastases from cancer of the breast, a quarter of the anterior lobe was occupied by an enormous adenoma, which had the histologic appearance described for malignant chromophobic hypophyseal adenoma. Even this tumor did not leave any mark on the habitus of the patient. Little is known about the fate of hypophyseal adenomas. I am able to show you a large adenoma which is almost completely hyalinized and has calcified masses in its center. Only small epithelial trabeculae are left.

Probably, if one made serial sections of the hypophysis as a routine, the percentage of adenomas would increase. So far one knows of no clinical effect of these tumors.

#### SOME OBSERVATIONS ON THE MORPHOLOGY OF GLANDULAR HYPERPLASIA OF THE PROSTATE. ROBERT A. MOORE.

With senility, certain morphologic changes occur in the prostate, apparently independent of any pathologic change. These consist of atrophy of the glands and fibrosis of the stroma. They have been given in greater detail in a previous paper. During this process of senile atrophy several types of hyperplasia may occur. For convenience they may be divided into nonnodular and nodular. In the nonnodular type the glandular hyperplasia may occur in focal areas, involving from two to ten acini, or may occur diffusely with involvement of all the acini. The nodular type is the well known hypertrophy of the prostate which involves both the glands and the stroma. It may arise at any point in the prostate, but is most common in the lateral lobes. Many nodules have no relation to the periurethral glands. Morphologically an active and an inactive stage of the hyperplasia may be distinguished. The incidence of the disease increases with increasing age to 75 per cent of all prostates in the ninth decade of life. Carcinoma rarely arises in these nodules of hyperplasia. Experimental studies are now being conducted in an attempt to relate the various types of hyperplasia to the altered morphologic and physiologic state of senility.

## Book Reviews

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**A Textbook of Histology: Functional Significance of Cells and Inter-cellular Substances.** By E. V. Cowdry, Professor of Cytology, Washington University, School of Medicine, St. Louis. Cloth. Price, \$5.50. Pp. 503, with 242 illustrations. Philadelphia: Lea & Febiger, 1934.

This book is an interesting departure from the conventional type of textbook of histology with the main emphasis on the detailed description of the microscopic structure of all parts of the body. The principal aim of the book is to build up a clear conception of the relation of minute structure and function in the organs and systems of the body through a well directed study of their constituent cells and elementary tissues. Details that for various reasons are considered as not essential to the immediate purpose have been omitted, e. g., illustrations of what the author calls the obvious, that is, figures showing "at a glance what the actual preparations will lead the students to discover for themselves." But there is no lack of good illustrations and diagrams of facts that are difficult for students to unearth. Many of these illustrations are original, while others are taken from the writings of the author, works on cytology edited by him and from other sources. A commendable feature of the book is the frequent use of tables, twenty-six in all, to present various data such as the nomenclature of the blood cells, the differential properties of the blood vessels, a comparison of the suprarenal cortex and medulla, the characteristics of the constituent parts of the hypophysis and chemical integration by hormonic action. In the latter list commercial names have been omitted almost completely. The effort to avoid undue repetition of the same information in the text, tables and illustrations is another commendable feature that should be copied widely, especially in articles in scientific medical periodicals. Enumeration of the subjects will give a good idea of the general plan and scope of the book: water, the essential vital medium (the body fluids); the blood, the principal integrator; absorptive drainage into the blood (lymphatics and spleen); chemical integration by endocrine products in the blood stream (ductless glands); intake of water, nutriment, accessory food factors and removal of waste (the digestive system); oxygen consumption and carbon dioxide elimination (respiratory system); regulation of constitution of blood and removal of waste (urinary system); rapid neural integration of response to internal and external stimuli (nervous system, sense organs); architectural support (connective system); movement (muscular system); perpetuation of the race (reproductive systems), and unification, protection and adjustment (skin). At the end of each chapter is a clear, concrete summary of the contents as illustrated by that on the spleen: "The spleen is the largest lymphoid organ in the body, but it contains no special cells not found elsewhere and hence can be removed without fatal results. In it the myriads of lymphocytes are not confined in vessels or nodes of their own, as in the lymphatic system proper; nor do they invade an epithelial rudiment, as in the thymus; but they meet the fluids and cells of the blood in the tissue fluid. To facilitate this unique association the walls of the capillaries and venules are made very thin and may even have openings. The circulation is so sluggish that the spleen is supplied with penetrating bands of smooth muscle, the shortening of which presses the blood onward. The chances for absorption by lymphocytes, exposed reticular cells of the connective tissue framework and vascular endothelial cells of substances in the blood stream are excellent. The spleen is indeed a great blood filter and the headquarters of the reticulo-endothelial system. The latter disposes of many sorts of particulate matter, including broken down red cells, and plays an important rôle in some immunological reactions. The spleen is also a blood reservoir capable by contraction of adding materially to the circulating blood. In response to special demands it may revert to its embryonic habit of

forming blood cells." At the end are a valuable bibliography and an index of the authors cited in the text. The style is clear, direct and vivid. Any tendency to rambling or prolixity is checked at once. The book meets admirably the goal held in view by its author "that the students shall gradually visualize more and more accurately the wonderful reactivity of the minute structure of the human body in terms of biochemistry, physiology and pathology and, finally, in practice shall institute measures of assistance with due caution." It will be welcomed by all who are interested in following the growth of knowledge of human microscopic structure in relation to function.

**Physiology in Health and Disease.** By Carl J. Wiggers, M.D., Professor of Physiology in Western Reserve University School of Medicine, Cleveland. Price, \$9. Pp. 1184, with 182 engravings. Philadelphia: Lea & Febiger, 1934.

This book was written to meet the needs of earnest students of medicine both before and after graduation from medical school. The author became impressed with the need of correlating his teachings as a physiologist as closely as possible with the efforts of his clinical colleagues to understand functional disturbances in disease. He found himself hampered in meeting this need in a practical way by the lack of a suitable textbook in which the essentials of physiology in health and disease are presented in suitable form. Herein lies the reason for this book. It is divided into ten sections in which the following subjects are treated: physiology of movement; the physiology of the peripheral and central nervous systems; the coordination of visceral function; the blood and blood-forming organs; respiration; the heart and circulation; the physiology of the alimentary tract; water transport and the excretory systems; metabolism and nutrition, and the endocrine organs and reproduction. The main subdivisions under each section are presented in separate short chapters, an arrangement of advantage for the casual reader as well as for supplementary study in connection with conferences and lectures, as the sequence of chapters may be changed as desired. At the end of each chapter is a selected list of appropriate monographs and reviews, while footnotes give references to articles, mostly recent, on particular topics. The book contains in well organized and well written form a wealth of information of the greatest importance to the study and understanding of physiology in its fundamental relations to modern clinical medicine. The author has rendered a great service to medical study.

**Pathologie und Klinik der Granulosazelltumoren.** By Dr. Walter Schiller, Assistent der II. Universitäts-Frauenklinik (Prof. Dr. Wilhelm Weibel), Vienna. Price, 16 marks. Pp. 197, with 136 illustrations. Vienna: Wilhelm Maudrich, 1934.

This is a review of granulation cell tumors based on study of original cases and of the literature. After a thorough consideration of the structure, histogenesis and clinical characteristics of the granulation tumors, the author comes to the general conclusion that they arise as primarily benign tumors from undifferentiated remnants of the mesenchymatous nucleus of the ovary. According to the grade of differentiation, they assume immature, trabecular forms or mature, folliculoid forms. In the majority of cases these tumors are unilateral. By the production of hormone they induce endometrial hyperplasia, which before the menopause may result in amenorrhea and eventually metrorrhagia, and after the menopause in irregular bleedings. The possibility of malignant granulation cell tumors must be granted, but it is not possible to separate them with complete certainty from genuine, primary, secondarily solid carcinoma of the ovary. Because the trabecular form of granulation cell tumor is not malignant, there is no justification to treat younger women after removal of such tumors so intensively with the roentgen rays as to produce unneeded castration. The book will interest all who wish to understand fully the granulation cell tumor. Unfortunately, it lacks an adequate index of subjects.

**A Practical Treatise on Diseases of the Skin for the Use of Students and Practitioners.** By Oliver S. Ormsby, M.D., Clinical Professor and Chairman of the Department of Dermatology, Rush Medical College, University of Chicago. With revision of the histopathology by Clark Wylie Finnerud, B.S., M.D., Assistant Clinical Professor of Dermatology, Rush Medical College, University of Chicago. Fourth edition. Cloth. Price, \$11.50. Pp. 1,288, with 622 illustrations. Philadelphia: Lea & Febiger, 1934.

This standard work appears in its fourth edition after thorough revision and with much new material. "Thirty-six new diseases are described, twenty rewritten, and the entire work brought up to date." The literature has been reviewed in an earnest effort to make the book reflect the knowledge of dermatology adequately. The references to its literature are limited largely to articles that contain complete bibliographies. The presentation is clear, concise and orderly. The author is a master in his field and writes under the full sense of his responsibility and authority as a teacher. A phenomenal grasp of the various phases of diseases of the skin is evident on every page. The book is well illustrated, and the pathologist will find the descriptions and illustrations of great help in understanding the structure of cutaneous lesions. The work of the publisher has been done very well.

## Books Received

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MEDICAL USES OF RADIUM. SUMMARY OF REPORTS FROM RESEARCH CENTRES FOR 1933. Medical Research Council, Special Report Series, No. 197. Price, 9 d. Pp. 40, with 4 illustrations. London: His Majesty's Stationery Office, 1934.

THE INHERITANCE OF RESISTANCE TO BACTERIAL INFECTION IN ANIMAL SPECIES; A REVIEW OF THE PUBLISHED EXPERIMENTAL DATA. A. Bradford Hill. Medical Research Council, Special Report Series, No. 196. Price, 1s. 3d. Pp. 71. London: His Majesty's Stationery Office, 1934.

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